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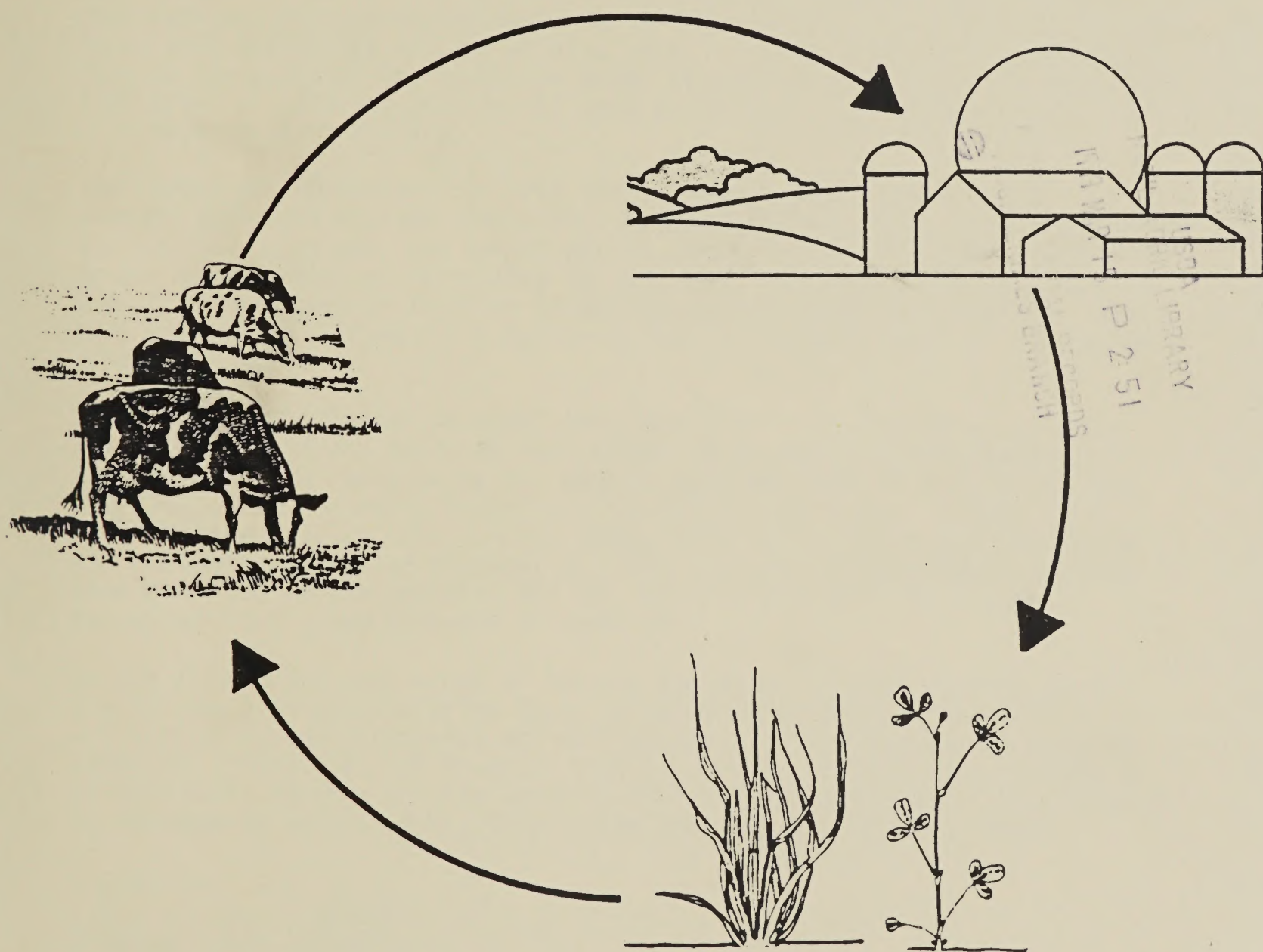
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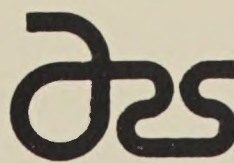# US DAIRY FORAGE RESEARCH CENTER

1986 RESEARCH SUMMARIES

U. S. Dairy Forage Research Center
1925 Linden Drive West
Madison, Wisconsin 53706

ars Agricultural Research Service

United States Department of Agriculture

U.S. DAIRY FORAGE RESEARCH CENTER, ARS-USDA
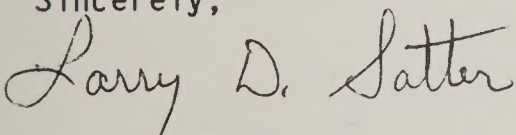Madison, WI 53706

Dear Reader:

It is a pleasure to update our progress by bringing you these summaries of
recent research. The U.S. Dairy Forage Research Center is a unique part of
the national research program of the Agricultural Research Service, U.S.
Department of Agriculture. The Center's mission is to build a knowledge and
technology base for the dairy industry to fully exploit the use of forages in
the production of milk. The Center has agricultural engineers, plant and soil
scientists, and ruminant nutritionists working together to increase the
efficiency of forage utilization by dairy farmers. We function in close
cooperation with the Agricultural Experiment Stations of several states. The
Center is located on the campus of the University of Wisconsin, Madison and
has "Cluster" locations in St. Paul, MN, Ames, IA, Columbia, MO, Wooster, OH,
East Lansing, MI, University Park, PA, and Ithaca, NY. The Center's research
farm, with facilities for 300 milking cows, is located on 63 acres of USDA
land on the banks of the Wisconsin River in Prairie du Sac, WI. An additional
1300 acres of adjacent land is utilized by the Center by agreement with the
U.S. Department of the Army.

The Center was established in 1980 and has made steady growth since. At
present there are sixteen scientists; eight at Madison, and one each at six of
the Cluster locations, and two at the St. Paul, Minnesota Cluster location.
We are in the midst of recruiting for a Chemist and a Microbiologist and
expect to have them on location in Madison within three to six months. We
will soon be recruiting for a person to fill a temporary position in the area
of Systems Analysis.

We have had some changes in staff this past year. Dr. Fred Ehle left the
Center program at St. Paul, MN last summer to go into a dairy consulting
practice. He was replaced by Dr. Hans G. Jung who came to us from the ARS,
U.S. Meat Animal Research Center in Clay Center, Nebraska. In what was a
surprise to all of us, Dr. Charles Kiddy, Director of the Center, announced
his retirement effective December 31, 1986. We were very sorry to see him
step down, but we are grateful for the leadership he gave the Center.
Recruitment for a new Director is under way.

We are pleased and very proud of the way the Center scientists from diverse
disciplines interact and bring their collective insights to bear on the
problems of forage production and utilization. This collection of research
summaries illustrates the progress they are making in developing information
to help dairy farmers utilize their resources more effectively. The research
is intended to benefit dairy farmers and the consumers of dairy foods.

Sincerely,

Larry D. Satter

Larry D. Satter
Acting Director
U.S. Dairy Forage Research Center

U.S. DAIRY FORAGE RESEARCH CENTER, ARS-USDA
Madison, WI 53706

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CONTENTS

FORAGE DEVELOPMENT

FORAGE EVALUATION

UTILIZATION OF CALCIUM AND PHOSPHATE FROM ALFALFA AND FESCUE BY LACTATING AND DRY DAIRY COWS

F.A. MARTZ, M. WEISS, R.L. BELYEA AND K.S. WALLER

INTRODUCTION

Data pertaining to the true absorption of calcium and phosphorus for dry and lactating dairy cows are scarce. Due to this lack of information the National Research Council (NRC) uses a common value of 45% for calcium and 55% for phosphorus for all feedstuffs. An improved understanding of true absorption of calcium and phosphorus for various feedstuffs will allow nutritionists to balance dairy cattle rations based on mineral availability and will thus result in dairy herds which are more healthy and which attain greater longevity.

The objectives of this research were to determine the true absorption of calcium and phosphorus from alfalfa and tall fescue forages when fed in a total mixed ration to high producing cows (greater than 27 kg milk/d) and to assess the utility and value of isotope tracer techniques for this type of study.

MATERIALS AND METHODS

Eight Holstein cows were paired by milk potential and parity then assigned to either a fescue or an alfalfa based diet. Mineral availability measurements were made during 14 d intensive periods of metabolic balance determination during the dry (28 $\pm$ 16 d), early (64 $\pm$ 16 d) and post-peak (155 $\pm$ 16 d) stages of a lactation cycle and were statistically analyzed as subplots in the split plot in time design. Determination of true absorption was by comparative balance technique, intravenous doses of Ca-45 and P-32 vs. feeding intrinsically Ca-45 and P-32 labeled tracer alfalfa or fescue. Diets fed during lactation were composed of 24% of the base forage, complemented with corn silage and a grain mix. The A based diets received no Ca supplementation but included monosodium phosphate in order to narrow the Ca:P ratio to 1.5-2.0:1. Intrinsically labeled forage plants were produced by a hydroponic growing method where the Ca-45 and P-32 was introduced into the nutrient solution several times prior to harvest over a 14-day period.

RESULTS AND DICSUSSION

Milk production for the cows during the period of total collection is presented in Table 1 and intake of total mixed ration dry matter by the cows is presented in Table 2. As would be expected, cows consumed less dry matter when dry than when lactating. Cows consumed about 3.5% of their body weight as total mixed ration dry matter.

The calcium and phosphorus content and intake is illustrated in Table 3. The data indicate that due to the relatively high content of calcium in alfalfa, the dry cow ration containing alfalfa exceeded the level set by NRC for calcium. Also, the calcium content of the alfalfa ration exceeded that of fescue in all instances. Phosphorus content was more nearly equal for both rations except for the dry period where alfalfa again exceeded fescue.

Calcium intake from the alfalfa rations exceeded the fescue rations

which was a reflection of both ration dry matter intake and calcium content. Phosphorus intake was more nearly equal for both rations.

True absorption of calcium and phosphorus from the different forages is presented in Table 4. When the data are presented on this basis, it appears that calcium absorption increased from dry to early lactation to mid-lactation for alfalfa and that calcium absorption from fescue decreased. Examination of the data indicated that all cows were over fed calcium when compared to NRC requirements while consuming both alfalfa and fescue. When the calcium absorption was regressed on total calcium intake as a % of NRC requirements, the intercept for both alfalfa and fescue was about 40% which is about 5 percentage units less than the value of 45% used for NRC standards.

Similar analysis of the true absorption of phosphorus indicated that it did change with period or level of phosphorus consumed. True absorption was about 60 to 65% for both alfalfa and fescue which is about 10 percentage units greater than the NRC value of 55%.

These data indicate that the true absorption of calcium from alfalfa and fescue hay, when fed at levels of 25% of the ration and at NRC recommended levels, is about 40%. The true absorption of phosphorus from these forages, under these conditions, was about 60%. True absorption of calcium and phosphorus did not change significantly from day 60 to day 150 of lactation.

Table 1. Milk production during total collection periods.

Stage of Lactation	Alfalfa (Kg/d)	Fescue	$\overline{X}$
Early (60d)	34.6[a,c]	28.6[b,c]	31.6[e]
Middle (150d)	28.7[b,d]	25.8[b,d]	27.3[f]
$\overline{X}$	31.7[a]	27.2[b]	

[a,b] Denotes Signif. diff. in same row at P<.25
[c,d] Denotes signif. diff. in same column at P<.05
[e,f] Denotes signif. diff. in same column at P<.01

Table 2. Dry matter intake of cows during collection periods at three stages of lactation cycle.

Stage of Lactation	Dry Matter Intake	
	Alfalfa	Fescue
	(Kg/d)	
Dry	10.4[c]	9.5[c]
Early (60d)	21.8[c]	18.9[d]
Middle (150d)	21.1[c]	20.2[c]
$\overline{X}$	17.8[a]	16.2[b]

[a,b] Denote signif. diff in same row at P<.25
[c,d] Denote signif. diff in same row at P<.10

Table 3. Calcium and phosphorus content of ration and daily intake for cows consuming an alfalfa and fescue diet at three different lactation stages.

Stage of Lactation	Content of Feed				Mineral Intake			
	Alfalfa		Fescue		Alfalfa		Fescue	
	Ca	P	Ca	P	Ca	P	Ca	P
	(% DM)				(g/d)			
Dry	.97	.42	.49	.27	101	44	47	26
Early (60d)	.65	.39	.59	.47	141	85	111	89
Middle (150d)	.58	.39	.56	.44	122	82	113	89

Table 4. True absorption of 45Ca and 32P from labeled forage.

	45Ca		32P	
	Alfalfa	Fescue	Alfalfa	Fescue
	(%)		(%)	
Dry	25.4[c,f]	42.9[c,g]	55.6[j,m]	94.4[n]
Early (60d)	28.1[c,e,f]	37.0[c,g]	62.8[j,l,m]	66.1[m]
Middle (150d)	32.8[d,e]	36.8[c,f]	65.9[k,l,m]	67.3[m]
$\bar{X}$	28.8[a]	38.9[b]	61.4[h]	75.9[i]

a,b   Denote signif. diff. in same row at P<.05
c,d,e Denote signif. diff. in same column at P<.10
f,g   Denote signif. diff. in same row at P<.10
h,i   Denote signif. diff. in same row at P<.01
j,k,l Denote signif. diff. in same column at P<.05
m,n   Denote signif. diff. in same row at P<.05

EFFECT OF ALFALFA MATURITY ON MILK PRODUCTION, DIGESTIBILITY, AND RATE OF PASSAGE IN DAIRY CATTLE

W.F. NELSON AND L.D. SATTER

INTRODUCTION

Increasing maturity of alfalfa reduces milk production in high producing cows. The extent to which milk production is reduced with increased maturity is not clear. This effect needs to be quantitatively examined so more efficient management of the dairy and forage enterprise can result. The objective of this experiment was to quantify the effect of increasing maturity of alfalfa on the performance of lactating dairy cattle at various levels of production.

METHODS AND PROCEDURES

First cutting Flemish varieties of alfalfa were harvested at three stages of maturity (mid bud, early flower, and late flower) and stored in separate concrete stave silos. A profile of the three maturities is presented in Table 1.

Fifty-five multiparous Holstein cows, ranging from 40-200 days in milk and 16-42 kg milk/day, were assigned to one of three treatment diets.

Three diets were formulated using the three maturities of alfalfa. In all three diets the alfalfa silage comprised 55% of the diet dry matter. Diets were fed twice daily as total mixed rations. All three diets were balanced to provide 17% crude protein using soybean meal and high moisture ear corn.

All 55 cows received the Early Cut diet for a three week pretrial period. Based on production in the pretrial period, cows were blocked into one of three production groups and randomly assigned within groups to either the Early, Mid, or Late Cut diet for a 10 week trial period. The final week of the pretrial period was used for covariate adjustment.

Feed and feed refusals were recorded and sampled daily. Body weights were recorded once weekly. Daily milk production was recorded daily and milk samples were taken once weekly at morning and evening milkings.

Diet digestibility was measured using rare earth elements as markers during week 10 of the trial period using all cows. Rate of passage was measured on the high production groups during week 10.

RESULTS AND CONCLUSIONS

Maturity of alfalfa had the greatest influence on the high production group, reducing milk yield by approximately 4 kg milk/day when going from mid bud to full flower alfalfa (Table 2). The 27-day difference in cutting date of these two maturities results in a potential loss of .15 kg of milk for each day harvest is delayed past the mid bud stage of maturity for cows averaging 30 kg milk/day.

Earlier research (Kawas and Jorgensen), using alfalfa hay as a forage source, showed that the cost of delayed harvest may be as much as .45 kg milk per day of delay in harvest with cows producing in excess of 40 kg per day. A linear extrapolation of results from our

study indicates that this value is closer to .23 kg milk/cow/day per day of delay in harvest for the 40 kg cow. It is clear that an increase in maturity has much less detrimental effect on mid and late lactation cows than on early lactation cows.

Diet DMD decreased with increasing maturity of alfalfa and a slight increase in rumen retention time (RRT) was seen with full flower alfalfa relative to early and mid cut alfalfa.

Table 1. Forage maturity and composition.

	Date Cut	Mean Stage[a] by Weight	% CP	% NDF	% ADF
Early cut (Early bud)	5/30	2.5	20.9	40.2	36.0
Mid cut (Late bud)	6/14	4.3	18.6	52.0	44.4
Late cut (Late flower)	6/27	6.2	17.0	54.9	47.6

[a] Kalu and Fick. Crop Science 21:267.

Table 2.   Effect of maturity on milk production, feed intake, digestibility and rumen retention time.

	HIGH GROUP			MID GROUP			LOW GROUP		
	E	M	L	E	M	L	E	M	L
Milk, kg/d	30.3	28.8	26.2	23.2	24.1	23.7	17.7	18.2	16.0
4% FCM, kg/d	30.6	29.3	26.6	25.1	24.7	23.8	19.2	18.3	16.8
Milk fat, %	4.08	4.20	4.05	4.35	4.10	4.09	4.36	4.08	4.43
Milk protein,%	3.32	3.29	3.28	3.38	3.25	3.37	3.62	3.61	3.50
DMI, kg/d	25.3	24.9	23.6	23.8	22.9	22.2	19.8	20.3	18.5
DIM, d	141	142	139	153	157	136	202	201	209
DMD, %	66.6	66.5	61.6	66.9	64.1	64.5	66.5	63.6	63.1
RRT, h	16.8	16.3	17.7	---	---	---	---	---	---

E = early cut diet   FCM = fat corrected milk   DMD = dry matter digestibility
M = mid cut diet   DMI = dry matter intake   RRT = rumen retention time
L = late cut diet   DIM = days in milk

EFFECT OF METHOD OF PRESERVATION AND MATURITY
OF ALFALFA ON MILK PRODUCTION, DIGESTIBILITY
AND RATE OF PASSAGE IN DAIRY CATTLE

W.F. NELSON AND L.D. SATTER

INTRODUCTION

The effect of maturity of alfalfa on milk production was measured in an earlier trial. Feeding more mature alfalfa reduced milk production in early lactation cows but had little effect with mid and late lactation cows. Based on literature information, a greater reduction in milk production would have been expected. Since alfalfa was fed as silage in our experiments, and as hay in most of the literature studies, this experiment was designed to study the interaction of forage maturity and method of preservation.

METHODS AND PROCEDURES

Three maturities of first cutting alfalfa, similar to those in an earlier trial, were harvested in 1985. Half of each maturity was conserved as dry hay and half was conserved as silage. Table 1 contains a profile of the alfalfa silage and hay.

Eighty-five multiparous Holstein cows ranging from 40 to 180 days in milk and 18 to 45 kg milk/day were assigned to one of three treatment diets.

Forty-four cows were on experiment in the fall of 1985 and 41 cows on experiment in the spring of 1986. Methods used were similar to those used in an earlier trial with the following exceptions:

The Early Cut silage diet was fed to all animals for a two week pretrial period with the last week used for covariate adjustment. Cows were not blocked by production. Silage diets were fed as total mixed rations once daily. Cows on hay diets were fed hay once daily and grain twice daily. All diets consisted of 55% forage:45% grain (dry matter basis) and were balanced to provide 19% crude protein. Dry matter digestibility (DMD) and rate of passage were determined for all cows during week 10 of the fall trial and week 5 of the spring trial.

RESULTS AND CONCLUSIONS

Milk production was not depressed due to increasing alfalfa maturity with the silage diets (Table 2). The mid cut and late cut hay diets depressed milk production relative to the early cut hay diet. Across all three stages of maturity, the hay diets resulted in lower milk production than the silage diets. DMD decreased with increasing maturity with both hay and silage. DMD was lower for the silage diets across all three maturities due to shorter rumen retention times (RRT) for the silage diets. There was no increase in RRT with increasing maturity in silage diets. However, increased maturity of the hay diets tended to increase RRT.

Linear extrapolations of the data from this trial indicate that there was little or no depression in milk production associated with increasing maturity of alfalfa conserved as silage for the 40 kg cow. If alfalfa was conserved as hay, however, a potential depression of approximately .23 kg/cow/day per day of delayed harvest exists. More

research is needed to determine why preservation of alfalfa as silage compared to hay results in less detrimental effects on milk production as alfalfa maturity increases.

Table 1. Forage maturity and composition.

	Date Cut	Mean Stage[a] by Weight	% CP	% NDF	% ADF	% ADL
Early cut silage (Early bud)	5/18	2.5	21.5	41.7	33.7	7.7
Mid cut silage (Late bud)	6/1	4.0	19.3	48.8	39.1	8.5
Late cut silage (Late flower)	6/13	6.0	18.0	51.5	39.5	8.9
Early cut hay (Early bud)	5/17	2.5	21.0	42.4	32.5	8.0
Mid cut hay (Late bud)	6/1	4.0	17.9	50.2	38.8	8.3
Late cut hay (Late flower)	6/13	6.0	16.5	52.3	40.1	8.8

[a] Kalu and Fick. Crop Science 21:267.

Table 2. Effect of method of preservation and maturity on milk production, feed intake, digestibility and retention time.

	SILAGE			HAY		
	Early	Mid	Late	Early	Mid	Late
Milk, kg/d	27.2	27.0	27.7	26.8	25.6	25.6
4% fat corrected milk, kg/d	26.8	26.2	27.5	25.1	24.4	24.6
Milk fat, %	3.95	3.82	3.97	3.59	3.78	3.74
Milk protein, %	3.31	3.21	3.25	3.20	3.13	3.13
Dry matter intake, kg/d	22.2	22.4	22.6	20.2	18.8	18.1
Dry matter intake, %BW	3.60	3.63	3.72	3.45	3.26	3.36
Days in milk, d	145	141	141	145	139	13.8
Dry matter digestibility,%	62.2	61.3	57.2	65.4	61.6	60.2
Rumen retention time, h	15.5	16.1	16.2	20.5	21.8	23.9

FEEDING LACTATING DAIRY COWS PROTEINS WHICH ARE RESISTANT TO RUMINAL DEGRADATION

V.L. VOSS, L.D. SATTER AND G.A. BRODERICK

INTRODUCTION

Degradation in the rumen of protein in alfalfa silage may be greater than with protein in corn silage. Most of the published work with resistant proteins has been with diets containing corn silage as the only forage. The response to resistant proteins may be different with alfalfa silage. The objective of this study was to measure the milk production response to supplementation of resistant protein sources with diets containing either a corn silage-alfalfa silage combination or alfalfa silage as the sole forage. Heated soybean meal and soybeans and a combination of distillers dried grains and corn gluten meal were the resistant protein sources used.

METHODS AND PROCEDURES

One hundred and five multiparous Holstein cows were fed a pretreatment diet from days 4-13 postpartum (covariate period) and then were placed on treatment diets for 60 days. Treatments varied with respect to supplemental protein source and forage. Treatments 1-5 contained equal amounts of corn silage and alfalfa silage as a forage source and contained either solvent soybean meal, roasted soybean meal, roasted soybeans, roasted soybeans and urea, or a mixture of corn distillers dried grains and corn gluten meal as the supplemental protein. Treatments 6 and 7 had alfalfa silage as the forage source and either solvent soybean meal or roasted soybeans as the supplemental protein. All diets contained 55% forage:45% concentrate (dry matter basis). The concentrate consisted of high moisture ear corn, mineral and vitamin mix, plus one of the supplemental protein sources. Milk production was regressed on the milk production data averaged during the last five days of the covariate period. Comparisons between treatments were performed using orthogonal contrasts (Table 2).

RESULTS AND CONCLUSIONS

Feeding roasted soybeans increased milk 2.0 kg/day, 4% fat corrected milk 4.6 kg/day and milkfat .23 kg/day when compared to solvent soybean meal with alfalfa silage. No significant differences in milk production were found with diets containing corn silage-alfalfa silage when roasted soybean meal and roasted soybeans were compared with soybean meal and when roasted soybeans were compared to roasted soybean meal. Milk protein production was depressed with a combination of distillers dried grain and corn gluten meal when compared to diets with soybean sources with corn silage-alfalfa silage diets. Resistant protein sources may have greater value with diets containing alfalfa silage than diets containing corn silage.

Table 1. Means across all treatments for feed intake, body weight change
and milk production.

	Mean	Std. Deviation
Dry matter intake, kg/d	20.2	.45
Total body weight change, kg	-4.3	6.67
Milk, kg/d	37.1	.72
4% FCM, kg/d	35.0	1.02
Milk fat, %	3.63	.11
Milk protein, %	2.81	.04
Kg feed/kg milk	.55	.016

Table 2. Orthogonal Contrasts

Contrasts	Parameters						
	Dry Matter Intake (kg/d)	Total Body Weight Change (kg)	Milk (kg/d)	4% FCM (kg/d)	Milk Fat (%)	Milk Protein (%)	kg feed kg/Milk
Mixed silage vs. alfalfa silage							
Estimated difference[1]	-.24	-4.8	-.91	-1.6	.03	.06	.02
Probability level	.56	.43	.16	.08	.80	.11	.22
Solvent soybean meal vs. roasted soybeans with alfalfa silage							
Estimated difference[2]	1.35	-4.8	2.0	4.6	.36	-.12	-.01
Probability level	.05	.64	.07	.004	.05	.05	.69
Soybean sources vs. CDG-CGM with mixed diets							
Estimated difference[3]	-.004	.42	-1.0	-.9	.19	-.01	.03
Probability level	.99	.96	.26	.47	.17	.84	.14
Solvent soybean meal vs. roasted soybean meal and roasted soybean diets							
Estimated difference[4]	.06	2.1	-.2	1.5	.06	-.09	-.02
Probability level	.91	.80	.87	.24	.66	.07	.31

1   Estimated difference = mean for alfalfa silage - mean for mixed silage.
2   Estimated difference = mean for roasted soybeans - mean for solvent
    soybean meal with alfalfa silage.
3   Estimated difference = mean for corn distillers grain and corn gluten
    meal (CDG-CGM) - soybean sources with mixed diets.
4   Estimated difference = mean for roasted soybean meal and both roasted
    soybean diets - mean for both solvent soybean meal diets.

SUBSTITUTION OF DEHYDRATED ALFALFA WITH OR WITHOUT UREA FOR GRAIN IN DAIRY DIETS

S. PRICE AND L.D. SATTER

INTRODUCTION

Dehydrated alfalfa has traditionally been used to replace forage in lactating dairy cow diets. The possibility of dehydrated alfalfa substituting for grain, however, has not been fully examined. Substitution of dehydrated alfalfa (DEHY) for grain could enhance utilization of nonprotein nitrogen in the diet because of the relatively low degradation in the rumen of DEHY protein. Combination of a resistant protein source such as DEHY with nonprotein nitrogen might provide an economical substitute for protein.

METHODS AND PROCEDURES

This experiment consisted of two trials. Diets consisted of 50% corn silage and 50% grain mix and dehydrated alfalfa (DM basis) (Table 1). Dehydrated alfalfa replaced 40% of the grain in the DEHY and DEHY + urea diets. Diets were fed ad libitum 4X daily. The objective of Trial 1 was to measure ruminal parameters of lactating dairy cows fed either the SBM, DEHY, or DEHY + urea diets. The objective of the second trial was to measure milk production, milk composition, and plasma amino acids of early lactation dairy cows fed the same diets as Trial 1.

Trial 1 was an incomplete block design with two 14-d periods. Ruminal fluid was collected at 0, 1, 3 and 5 h post feeding from 6 ruminally cannulated cows in mid lactation. Analyses of pH, volatile fatty acids (VFA) and ammonia were conducted.

Trial 2 was a 3X3 Latin square design with 28-d periods using nine Holstein cows in early lactation. Feed consumption and milk production were recorded daily. Milk samples were collected on two consecutive d during each week of wk 3 and 4. Blood was drawn at 1 and 5 h post feeding on two consecutive days during the last week for plasma amino acid analysis.

RESULTS AND CONCLUSIONS

Total ruminal VFA concentration was increased ($P<0.05$) with the DEHY diet compared to the SBM diet (Table 2). Dry matter intake, milk production, and milk fat percentage for SBM, DEHY and DEHY + urea diets were: 24.2, 23.8, 24.0; 34.7, 33.4, 32.8; and 3.48, 3.58, 3.63, respectively. A trend toward increased milk fat test with the DEHY and DEHY + urea diets resulted in no significant differences in 4% fat corrected milk (FCM) between the three diets. Plasma concentrations of the branched chain amino acids, leucine, isoleucine, and valine increased by an average of 35% and 5% for the DEHY and DEHY + urea diets, respectively. This indicates an increase in total protein absorption compared to the SBM diet.

Table 1. Diet Ingredients

Ingredient	Soybean Meal	DEHY	DEHY + Urea
	% (DM Basis)		
Corn silage	50.1	50.0	50.0
Ground yellow corn	30.9	16.3	22.8
Dehydrated alfalfa	--	20.3	20.5
Soybean meal	17.7	12.8	5.1
Urea	--	--	0.9
Mineral and vitamin mix	1.3	0.6	0.7
Crude protein content	16.8	17.1	16.8

Table 2. Measurements

	Soybean Meal	DEHY	DEHY + Urea	$S_{\bar{x}}$
Rumen				
pH	6.18	6.02	6.06	.02
Total VFA (mM)	101 [a]	116 [b]	113 [ab]	1.70
Ammonia (mM)	6.03 [a]	5.34 [a]	8.05 [b]	.14
Animal Performance				
Dry Matter Intake (kg/d)	24.2	23.8	24.0	.15
Body Weight Change (kg/d)	0.53	0.25	0.15	.13
Milk Production (kg/d)	34.7 [a]	33.4 [b]	32.8 [b]	.3
Milk Fat (%)	3.48	3.58	3.63	.02
4% FCM (kg/d)	31.6	31.0	30.7	.2
Milk Protein (%)	3.13 [a]	3.00	2.98 [b]	.04
4% FCM/DMI	1.31	1.30	1.28	.01
Plasma Amino Acid Concentration (mM)				
Total amino acids[c]	874 [ab]	939 [b]	832 [a]	13.1
Essential amino acids[d]	361 [a]	450 [b]	364 [a]	10.1
Nonessential amino acids[e]	513 [a]	489 [ab]	468 [b]	5.5
Valine	96.6 [a]	133 [b]	101 [a]	3.2
Isoleucine	42.8 [a]	58.4 [b]	44.9 [a]	1.8
Leucine	62.2 [a]	81.9 [b]	67.1 [a]	2.7
Total Branched Chain Amino Acids[f]	202 [a]	273 [b]	213 [a]	7.8

a,b Means in the same row with different superscripts differ (P<.05).
 c   Included essential and nonessential amino acids as described below.
 d   Included Thr, Val, Met, Isol, Leu, Phe, Try, Lys, His, Arg.
 e   Included Asp, Ser, Asn, Glu, Gln, Pro, Gly, Ala, Cys, Tyr.
 f   Included Val, Ileu, Leu.

UTILIZATION OF DIETARY PROTEIN BY DAIRY COWS SUPPLEMENTED WITH SOYBEAN MEAL OR DEHYDRATED ALFALFA:CORN GLUTEN MEAL

S. PRICE AND L.D. SATTER

INTRODUCTION

Interest in formulating dairy diets to supply more undegraded dietary protein is increasing. The fractions of undegraded dietary protein in dehydrated alfalfa, corn gluten meal, and soybean meal have been estimated at 45, 55 and 30%, respectively. It appears that lysine and methionine are likely to be the first limiting amino acids for milk production. Dehydrated alfalfa, a rich source of lysine, and corn gluten meal, high in methionine, should be a good combination of protein supplements for meeting the amino acid requirements of lactating dairy cows fed corn-based diets. The objective of this study was to measure the utilization of dietary protein by lactating dairy cows supplemented with soybean meal or dehydrated alfalfa:corn gluten meal.

MATERIALS AND METHODS

Four cows fitted with ruminal cannulae and T-type cannulae in the proximal duodenum and terminal ileum were used in a switchback experiment. Diets consisting of 50% concentrate and 50% corn silage (DM basis) were fed ad libitum 4X daily. Diet ingredients are in Table 1.

Ruminal fluid was collected 2, 4, and 6 h post feeding. Duodenal, ileal, and fecal samples were obtained every even h of a 24-hour day over the last four days of each period. An additional six duodenal samples were obtained during those four days.

RESULTS AND CONCLUSIONS

No differences were observed for intakes of dry matter, organic matter, nitrogen, total amino acids, or essential amino acids (Table 2). Seventy-six percent of the dietary protein was degraded in the rumen of cows on the SBM diet compared to 62 percent with the Dehy:Gluten meal diet. Flow of total amino acids to the duodenum was 13% higher for the Dehy:Gluten meal diet than for the SBM diet. Apparent absorption of amino acids from the small intestine was 60% and 67% for the SBM and Dehy:Gluten meal diets, respectively. Protein utilization for the Dehy:Gluten meal diets was equal to or slightly better than the SBM diet under the conditions of this experiment.

Table 1. Diet Ingredients

Ingredient	Soybean Meal	Dehy: Corn Gluten
	% (DM Basis)	
Corn silage	52.9	52.2
Ground yellow corn	30.3	14.8
Soybean meal	15.6	--
Dehydrated alfalfa	--	25.3
Corn gluten meal	--	6.8
Mineral and vitamin mix	1.2	0.9

Table 2. Digesta Measurements

Measurement	Soybean Meal	DEHY:Corn Gluten Meal	$S_{\bar{x}}$
Rumen			
Ammonia (mM)	7.11 [a]	4.50 [b]	.1
Total VFA (mM)	104	99.2 [b]	1.2
Organic Matter			
Intake (kg/d)	15.8	16.0	.2
Flow to duodenum (kg/d)	8.95	9.82	.3
Flow to terminal ileum (kg/d)	6.95	7.61	.4
Fecal output (kg/d)	5.63	6.52	.2
Apparent digestibility in total gastrointestinal tract (%)	64.6	59.5	1.1
Nitrogen			
Intake (g/d)	386	382	6.2
Flow to duodenum (g/d)	376	426	15.9
Bacterial N	273	256	14.3
Dietary N[a]	103	170	19.8
Flow to terminal ileum (g/d)	154	163	8.7
Fecal output (g/d)	148	150	5.7
Apparent digestibility in total gastrointestinal tract (%)	61.6	60.9	1.0
Amino Acids[b]			
Intake (g/d)	2080	2160	82.4
Flow to duodenum (g/d)	1790	2060	83.0
Flow to terminal ileum (g/d)	717	676	29.2
Lysine			
Intake (g/d)	117.6 [a]	80.7 [b]	5.3
Flow to duodenum (g/d)	112.4	116.1	5.9
Methionine			
Intake (g/d)	29.0	27.2	1.7
Flow to duodenum (g/d)	28.6	42.8	3.5

[a] Includes endogenous and protozoal nitrogen.

[b] Includes: Arg, His, Ileu, Leu, Trp, Met, Phe, Thr, Val, Asp, Ser, Glu, Pro, Gly, Ala, Tyr

ALKALI-TREATED FORAGE IN COMPLETE RATIONS FOR DAIRY COWS: EFFECTS ON RUMEN DYNAMICS AND NUTRIENT DIGESTIBILITY

C.J. CANALE, S.M. ABRAMS, L.D. MULLER, W.L. KJELGAARD, AND P.M. ANDERSON

INTRODUCTION

Application of sodium hydroxide (NaOH) to low quality roughages is an established way to increase fiber digestibility in sheep and beef cattle. Research examining the treatment of medium and/or high quality forage is limited, and investigations describing utilization of such forage in early-lactation dairy cows is virtually nonexistent. Our objective was to determine nutrient digestibilities, rumen dynamics, and in situ digestion kinetics in such cows fed first-cutting alfalfa/ orchardgrass hay treated at mowing with NaOH. Hay composition, ration intake, and milk production were reported in last year's research summaries.

MATERIALS AND METHODS

A first-cutting mixture of alfalfa and orchardgrass was either untreated, or treated with 4 g NaOH/100 g forage dry matter at mowing and baled as hay. Hays were chopped to a particle length of one inch, fed in complete diets consisting of 55% hay and 45% concentrate (DM basis), and fed to eight early lactation Holstein cows (four with rumen cannulae) in a single reversal design. Each experimental period consisted of 28 d (12 d adaptation and 16 d data collection). Animals were fed ad libitum throughout the experiment.

On day 13 of each period, rumen liquid turnover was estimated using the liquid phase marker, Cobalt (Co) EDTA. Cows were dosed via rumen cannulae with 5 g Co EDTA in 200ml water 1 h prior to the morning feeding. Rumen fluid samples were taken every 2 h for 12 h post feeding. Rumen pH was measured at each sampling of rumen fluid. A 5 ml aliquot of rumen fluid was analyzed for concentrations of volatile fatty acids (VFA). Ration digestibility was estimated on days 20 through 25 of each period. All animals were fitted with urinary catheters. On days 26 through 28 of the second period, the rate and extent of NDF and DM degradation of hays were determined in situ. Duplicate polyester bags containing 8 g of untreated or treated hay were incubated in the rumen of cannulated animals for 12, 24, 36, 48, 60, and 72 h. After removal, bags were rinsed, dried at 60°C, weighed, and the residue analyzed for NDF concentration.

RESULTS AND DISCUSSION

Results are shown in Table 1. Chemical composition was virtually identical in control and NaOH rations. When fed the treated hay diet, cows had increased digestion of NDF, ADF, and hemicellulose. Rumen pH was lower and production of VFA higher in cows fed treated hay. In addition, concentrations of rumen acetate were greater and rumen isobutyrate lower when cows were fed treated hay. Sodium hydroxide treatment did not affect the number of other VFA or the turnover of rumen liquid. Incubated hays treated with NaOH had higher NDF digestible fractions and lower indigestible fractions compared to untreated incubated hays. The

degradation rate and lag time were not affected by treatment. These results, in combination with intake and milk production data, indicate that treatment of medium quality forages with compounds that increase utilization of forage fiber can be beneficial to the dairy cow.

Table 1. Diet composition and effect of alkali treatment on digestion, rumen dynamics, fermentation products, and in situ NDF degradation.

Variable	Control	NaOH
Ration Composition		
CP, % DM	18.5	18.5
NDF, % DM	44.4	44.2
ADF, % DM	27.4	27.3
Ca, % DM	0.70	0.70
P, % DM 0.41	0.40	
NE_l (Mcal/kg)	1.46	1.45
Apparent Digestion %		
Dry matter	68.86	71.92
Organic matter	69.21	72.03
Crude protein	77.25	77.58
Neutral detergent fiber	57.12	63.63*
Acid detergent fiber	58.25	62.93*
Hemicellulose	55.14	64.49*
Rumen pH 6.33	6.17**	
Rumen (liquid) volume (L)	59.96	67.14
Liquid T/O (%/h)	14.63	14.61
Fermentation Products		
Total VFA (uM)	105.53	111.94*
Concentration (uM/ml)		
Acetate	69.07	75.06***
Propionate	20.22	20.95
Butyrate	11.58	11.94
Isobutyrate	1.13	0.96*
Isovalerate	1.77	1.51
Acetate:Propionate	3.46	3.61
In situ NDF Kinetics		
Digestion fraction (%)	49.29	55.52***
Digestion rate (h^{-1})	.057	.068
Lag time (h)	2.91	3.68
Indigestible fraction (%)	41.67	32.38***

  * Treatment effect (P<.10).

 ** Treatment effect (P<.05).

*** Treatment effect (P<.01).

RUMINAL ESCAPE VALUE OF PROTEIN FROM EXPELLER SOYBEAN MEAL

G.A. BRODERICK, D.B. RICKER AND L.S. DRIVER

INTRODUCTION

Most soybean meal fed to livestock in the U.S. is produced using solvent extraction to remove the oil. An alternative method which generates considerable heat during oil removal is the expeller process (also called "old-process" soybean meal). This heating serves to protect the protein from breakdown by rumen microbes, and expeller soybean meal has a higher escape value than protein in conventional solvent soybean meal. We found previously that expeller soybean meal which is heated to a maximum of 163°C during processing had a ruminal escape value which was about 65% greater than solvent meal (that is, the protein value of 6% dietary expeller soybean meal equaled that of 10% dietary solvent soybean meal). Protein in alfalfa haylage is known to be used inefficiently for milk production, and protein supplements are needed for high producing cows. The purpose of these studies was to see if the greater resistance of expeller meal protein could be used to advantage in milking cows fed alfalfa haylage.

MATERIALS AND METHODS

Two trials, each with 20 multiparous Holstein cows, were conducted using a replicated 4X4 Latin square design. Cows in early lactation, averaging about 35 kg milk/day, were randomly assigned to diets after grouping in squares based on production. Periods were 21 days; data were analyzed from days 8-21. Diets were total mixed rations; the basal ration (diet A) contained [dry matter basis (DMB)]: 55% alfalfa haylage and 43% corn, plus 2% vitamins and minerals, and no supplemental soybean meal (SBM). SBM replaced corn DM in the other three diets as follows: B (4.0% solvent SBM), C (4.2% expeller SBM), and D (6.7% solvent SBM). Diets B and C had equal supplemental crude protein (CP), and diet D had about 65% more supplemental CP than diets B and C.

RESULTS AND DISCUSSION

Contents of DM, CP and neutral detergent fiber in alfalfa haylage were: 54.6, 20.9 and 43.5% (trial 1); and 29.9, 20.5 and 47.1% (trial 2). Ration CP contents were: 18.0, 19.1, 19.1 and 19.5% (trial 1); and 16.7, 17.9, 17.6 and 18.8% CP (trial 2), for diets A, B, C and D. Diets in trial 1 exceeded desired CP levels; DM intake (DMI) averaged 24.4 kg/d and no differences were observed in milk production. Results from trial 2 for diets A, B, C and D were (kg/d): 19.5^b, 20.4^a, 20.3^a and $19.9^{a,b}$ (DMI); 29.0^b, 30.4^a, 31.1^a and 30.7^a (milk); and $.85^b$, $.92^a$, $.94^a$ and $.92^a$ (milk protein) ($^{a,b}P<.05$). Production of fat and lactose was not influenced by diet. In trial 2, the basal ration (diet A) exceeded NRC protein requirement by 10%, but supplementation of either solvent or expeller soybean meal increased milk production by 1.4 or 2.1 kg/day. This confirms that protein in alfalfa haylage is poorly utilized for milk production. Somewhat greater production on expeller soybean meal suggests that cows fed alfalfa haylage may use the heat-protected protein in expeller soybean meal more efficiently than that in solvent soybean meal.

MICROBIAL PROTEIN CATABOLISM IN THE RUMEN
G.A. BRODERICK AND R.J. WALLACE

INTRODUCTION

Protein breakdown by ruminal microorganisms leads to the production of concentrations of ammonia which frequently exceed microbial growth requirements. The excess ammonia is absorbed through the rumen wall and eventually excreted as urea. Peptides are intermediates in the degradation of protein to ammonia as well as nutrients for growing microbes. There is in vitro and in vivo evidence that peptide metabolism may be a rate-limiting step in the breakdown of at least rapidly degraded proteins. It is possible that ruminal protein catabolism may be arrested at this point. Other research suggests that ruminal microbes may adapt their degradative activity to specific proteins and that protozoa may be important in protein degradation. These initial in vitro and in vivo experiments were conducted at the Rowett Research Institute.

MATERIALS AND METHODS

A fluorimetric method based on reaction with fluorescamine was adapted to assay uptake of individual peptides by mixed ruminal organisms. This method yielded 20 to 100 times greater fluorescence with peptides than with the constituent amino acids. The metabolism of 14 di- and oligopeptides containing only L-amino acids was investigated in strained rumen fluid diluted in anaerobic buffer. Hydrolysis by mixed rumen organisms of peptide p-nitroanilides was assessed using diazotization of the released p-nitroaniline.

Adaptation of microbial degradative activity was tested in sheep fed all their supplemental nitrogen as urea, casein or egg albumin (a soluble protein resistant to ruminal degradation). In another in vivo study, protease and deaminase activities and the metabolism of peptides were measured in rumen fluid from ciliate-free sheep and from sheep with a limited population of small entodinia. The same measurements were repeated following inoculation of the latter group with a more typical mixed ciliate population.

RESULTS AND DISCUSSION

Addition of glucose and dithiothreitol to mixed rumen microorganisms did not alter their uptake of di- or trialanine. Values of V_{max} and K_m for di- and trialanine uptake were 1.4 and 1.9 nmol/min per mg dry matter (DM), and .30 and .14 mM, respectively. Dipeptide uptake was .43 to .90 nmol/min per mg DM. There was a wider range of tripeptide uptake (.5 to 1.6 nmol/min per mg DM). Tri-, tetra- and pentaalanine were removed at 1.57, .93 and .71 nmol/min per mg DM; uptake of amino acid residues as alanine oligopeptides was two to three times more rapid than uptake as dialanine. These data suggest that peptides will accumulate in rumen fluid during hydrolysis of rapidly degraded proteins. Rates of hydrolysis of peptide p-nitroanilides were of similar magnitude to peptide uptake.

Feeding egg albumin had no effect on its rate of rumen hydrolysis, relative to casein. Greater numbers

of proteolytic rumen bacteria, mainly <u>Butyrivibrio</u> spp., were isolated from sheep fed albumin; relative albumin hydrolysis by these isolates was similar to that found for isolates from urea- and casein-fed sheep. It appears that rumen bacteria do not adapt their proteolytic activity to degrade resistant soluble proteins. Protease and dialanine uptake activities of mixed rumen microorganisms were not significantly influenced by protozoa. Trialanine uptake, leucine aminopeptidase, amino acid deaminase and trypsin-like protease activities were 70, 107, 73 and 91% higher with the limited population, and 72, 58, 64 and 55% higher when mixed protozoa were present. The data indicate protozoa have a major role in these metabolic processes in the rumen.

CHEMICAL, IN VITRO AND IN SITU EVALUATION OF HEAT-TREATED SOYBEAN PROTEINS

V.L. VOSS, G.A. BRODERICK AND L.D. SATTER

INTRODUCTION

Extensive ruminal degradation impairs utilization of feed proteins such as soybeans and soybean meal. Heating has been suggested as a method to increase the amount of soybean protein which escapes the rumen undegraded, but the optimal conditions (temperature and time of heating) remain to be established. Several chemical and biological methods have been used to assess relative value of heat-treatments. The purpose of these experiments was to apply solubility, in vitro and in situ tests to help identify the optimal degree of heating of raw soybeans and soybean meal for ruminal protection of the protein without reducing digestion in the small intestine.

MATERIALS AND METHODS

Raw, whole soybeans (SB) and solvent-extracted soybean meal (SBM) were heat-treated by roasting with a Gem Roaster and holding the materials at elevated temperatures for up to 3 h in covered, 227 Kg barrels. Initial and final temperatures at mid-points in the barrels were 120 and 110°C for SB and 116 and 110°C for SBM. Samples were removed from barrels at 0, .5, 1.0, 1.5, 2.0, 2.5 and 3.0 h, cooled and analyzed. Soybeans and SBM heated for 3 h were fed to lactating cows in a separate study. Solubility of nitrogen (N) was determined using McDougall's buffer, and "unavailable N" was measured using acid detergent insoluble N (ADIN). In vitro degradability was estimated using incubations of the proteins in strained ruminal fluid diluted with McDougall's buffer and containing 1.0 mM hydrazine to help obtain quantitative recovery of ammonia and amino acids. Incubations were conducted over 4 h,

with sampling every .5 h. In situ N degradation rates were estimated using N disappearance from dacron bags suspended in the rumens of cannulated cows. Standard washing and Kjeldahl methods were used to determine residual N. Finally, a "mobile bag" procedure was used to study intestinal digestibility of protein which escapes the rumen. In situ bags were removed from the rumen after 16h, then inserted via cannulae into the abomasum. Residual N in bags recovered in the feces was assumed to represent unavailable protein which escaped the rumen.

RESULTS AND DISCUSSION

Contents of ADIN and buffer N-solubility, as well as in vitro and in situ degradation rates, are in Table 1. No differences were

Table 1. Acid detergent insoluble N (ADIN), buffer N-solubility and in vitro and in situ degradation rates of roasted soybeans and soybean meal (SBM).[1]

Protein	Time held at temperature (h)	ADIN (%)	N-solubility (%)	Protein degradation rate (h^{-1})	
				In vitro (k_d)	In situ (c)
Raw soybeans	--	2.5	30.5	.092[a]	.174[a]
Roasted soybeans	0	--	10.5	.060[b]	.118[a]
	.5	2.9	4.9	.027[c]	.112[a]
	1.0	3.3	4.3	.020[c,d]	.114[a]
	1.5	3.8	3.6	.022[c,d]	--
	2.0	3.2	5.1	.024[c,d]	.110[a]
	2.5	3.7	4.1	.018[d]	--
	3.0	2.9	3.7	.017[d]	.090[a]
Solvent SBM	--	--	11.8	.091[a]	.069[a]
Roasted SBM	0	--	9.4	.088[a]	.036[b]
	.5	3.0	8.7	.056[b]	.018[c]
	1.0	2.9	7.4	.051[b]	.019[c]
	1.5	3.0	7.2	.049[b]	--
	2.0	3.1	5.3	.027[c,d]	.015[c]
	2.5	3.4	6.2	.031[c]	--
	3.0	3.1	4.9	.019[d]	.012[c]
Casein	—	0	100	.173	—
Bovine serum albumin	—	0	100	.042	—

[1] Values for ADIN, N-solubility and in vitro degradation rate are from soybeans and SBM roasted October, 1984. In situ degradation rates are from soybeans and SBM roasted September, 1985.

a,b,c,d Degradation rates having different superscripts differ (P<.05). Statistics were performed separately on soybeans and SBM groups. Casein and bovine serum albumin data are included for comparison.

detected in ADIN among the unheated and heated SB and SBM, suggesting that heating was not extensive enough to cause protein damage. The mobile bag technique also yielded no differences; overall N disappearances averaged 99.7% for SB and 99.0% for SBM. The degree of heating used in this experiment did not appear to result in over-protection of the protein.

Solubility of N in SB fell from 31 to 5% after .5 h and remained relatively constant up to 3 h of heating. However, buffer N-solubility for SBM continually declined with increased heating. Numerically, there were large differences between degradation rates observed for SB using the in vitro and in situ systems (Table 1). The pattern found with the in vitro system of decreased degradation rate with increased heating is largely absent from in situ data for SB protein. However, a similar decline of degradability with increased heating of SBM was observed with both in vitro and in situ methods. Degradation rates, for SB and SBM by in vitro and for SBM by in situ method, fell considerably with increased heating up to 2.0 h. Because degradation rates did not appear to plateau by 3 h, it is not certain that optimal protection has yet been reached. The large decreases in degradation observed here indicate that ruminal escape of SB and SBM heated for 2 to 3 h was substantially increased and suggest utilization by lactating cows would also be improved.

THE PRODUCTION OF TRICARBALLYLATE BY RUMINAL MICROORGANISMS AND ITS EFFECT ON ANIMAL METABOLISM

J.B. RUSSELL, R. SCHWARTZ AND H. MAYLAND

INTRODUCTION

Magnesium deficiency of ruminants has been recognized since the 1930's, and it is most often observed in cattle grazing succulent, cool season grass species. Grasses causing Mg deficiency and tetany often accumulate high concentrations of trans-aconitic acid, and it was hypothesized that this acid was chelating Mg and decreasing Mg availability. Created-wheat grass is the dominant western range species, and it can accumulate as much as 6% trans-aconitic acid. Cattle losses from Mg tetany were as high as 5% per year but Mg supplementation and management has decreased losses to less than 1%. This research describes the metabolism of trans-aconitic acid by rumen microorganisms, and the effect of its metabolite on animal metabolism.

MATERIALS AND METHODS

Mixed rumen bacteria were incubated with trans-aconitic acid and the resulting fermentation acids were separated and quantitated by high pressure liquid chromatography. An

unknown acid was subsequently identified as tricarballylate by chemical ionization mass spectrometry. Purified aconitase was assayed by an enzymatic method, and competitive inhibition by tricarballylate was noted on a Lineweaver-Burk plot. Isolated liver cells and slices were incubated with varying amounts of tricarballylate, and effects on ^{14}C acetate oxidation were used as an index of citric acid cycle inhibition. Blood samples from sheep dosed with trans-aconitic acid and sheep and cattle grazing wheat grass were assayed for tricarballylate as described above. Young rats were fed either tricarballylic or citric acid, and the Mg and Ca excretion was compared to controls not receiving acid.

RESULTS AND DISCUSSION

Mixed rumen bacteria converted more than 40% of the added trans-aconitic acid to tricarballylic acid (Figure 1) and when methane inhibitors were added the conversion was increased to 83% (Table 1). Selenomonas ruminantium, a common rumen bacterium, was the most active tricarballylate producer. Sheep fed trans-aconitic acid absorbed tricarballylic not trans-aconitic acid, into blood, and the concentration was greater than 0.5 mM. Cattle fed wheat forage had similar concentrations in blood even though the trans-aconitic acid content of the forage was not particularly high (approx. 1.5%). Tricarballylic acid was a competitive inhibitor of the citric acid cycle enzyme, aconitase, and the inhibitor constant, K_i, was 0.52 mM. The K_i was similar to the K_m of the enzyme 0.33 mM. Oxidation of ^{14}C-acetate by rat liver cells was inhibited by 30% when 0.5 mM tricarballylate was added to the incubation buffer. Rats fed 2% tricarballylic acid had 4 times greater Ca excretion and 2 times greater Mg excretion. Rats fed citric acid, a structurally similar but metabolizable acid, had cation excretions similar to controls. Virtually all of the dietary tricarballylate was recovered in the urine, and the recovery indicated that this acid could not be metabolized by the animal. Based on these results, tricarballylic acid could be a significant factor in the magnesium deficiency commonly known as grass tetany.
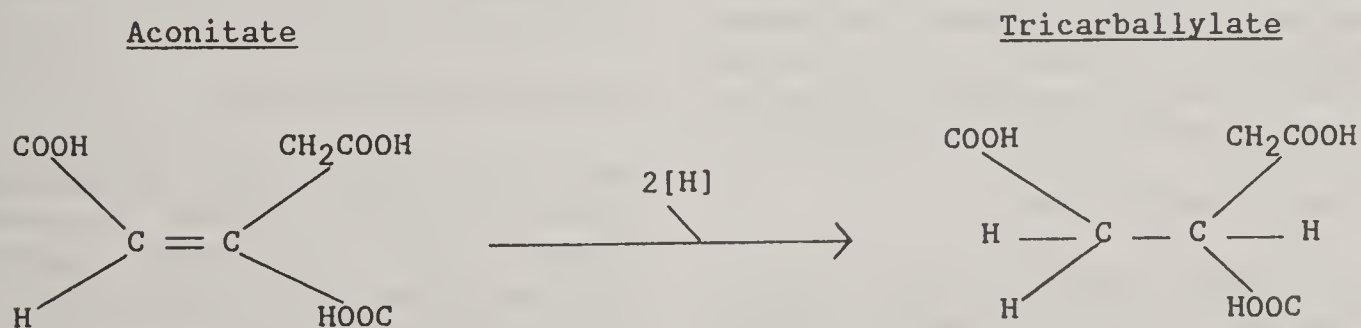
Aconitate

Tricarballylate

Figure 1. The reduction of trans-aconitate to tricarballylate.

Table 1. Effect of methane inhibition on the conversion of <u>trans</u>-aconitate to tricarballylate.

Additive	Methane (mM)	Tricarballylate Aconitate (%)
none	6.9	64
nitrate	2.8	76
chloroform	0.1	83

ENERGY SPILLING REACTIONS IN RUMEN MICROORGANISMS

J.B. RUSSELL AND H.J. STROBEL

INTRODUCTION

In ruminant animals, feedstuffs are fermented in the rumen prior to gastric and intestinal digestion. Since dietary proteins are often degraded to ammonia and volatile fatty acids, microbial protein represents a significant (and usually most important) amino acid source for ruminant animals. For many years it was assumed that yield of bacteria was relatively constant, but it now appears that growth rate and other environmental factors can have a significant impact on yield. This research examined the effect of carbohydrate accumulation and low pH on growth yields of rumen bacteria.

MATERIALS AND METHODS

Mixed rumen bacteria were grown in carbohydrate-limited batch cultures at pH 6.7 and 6.0. Microbial protein from washed cells was measured by the method of Lowry et al. (1951) while fermentation acids and carbohydrates were assayed by high pressure liquid chromatography. In other experiments, the heat production of <u>Bacteroides ruminicola</u> or <u>Selenomonas ruminatium</u> was measured in continuous culture with a microcalorimeter. Complete energy balances on these steady state cultures were derived from direct measurement of heat, bomb calorimetry of the cells, and the enthalpy of the fermentation acids.

RESULTS AND DISCUSSION

When the mixed rumen bacteria were grown at pH 6.0 (final pH greater than 5.7), microbial protein synthesis was 34 to 69% less than when the pH was 6.7. The reduction in protein was greater than decreases in carbohydrate

utilization, lactate accumulation and associated decreases in ATP (Table 1). Because less ATP was used for protein synthesis, it appeared that low pH directed energy to nongrowth or energy spilling reactions. Energy spilling was greatest when pectin, xylan, and cellobiose were the energy sources.

When the pure cultures were grown in continuous culture, heat production ranged from 2 to 34% of energy in glucose. At slow growth rates, the specific heat production (heat per unit cell mass) remained relatively constant, and the heat due to growth was small compared to the heat due to maintenance. Since heat of growth was small, total heat production provided a reasonable estimate of maintenance expenditures. As dilution rate and growth rate were increased, glucose eventually accumulated in the chemostat vessel. Glucose accumulation was associated with a more than twofold increase in specific heat production. Pulse doses of glucose into the glucose limited cultures likewise caused an immediate doubling of heat production and little increase in cell protein or mass (Figure 1). These experiments indicated that bacterial maintenance is not necessarily a constant and that energy can be diverted to heat production.

Further work is needed to determine the factors affecting the efficiency of rumen microbial protein synthesis. However, it is now clear that both low pH and carbohydrate accumulation can decrease protein synthesis and divert energy to nongrowth functions. The mechanisms involved in the energy spilling or heat production are not entirely true, but preliminary experiments would indicate that a futile cycle of ions through the cell membrane could be responsible.

Table 1. Analysis of metabolic effects leading to a decrease in rumen bacterial protein synthesis at low pH.

Substrate	Carbohydrate utilized	ATP[1] per carbohydrate	Bacterial protein	Energy spilling
	%-----------------------------------			
Starch	−15	−19	−35	1
Sucrose	−14	−19	−34	1
Cellobiose	−15	−23	−49	11
Xylan	−23	0	−37	14
Pectin	−53	0	−69	16
Mix[1]	−21	−1	−42	17

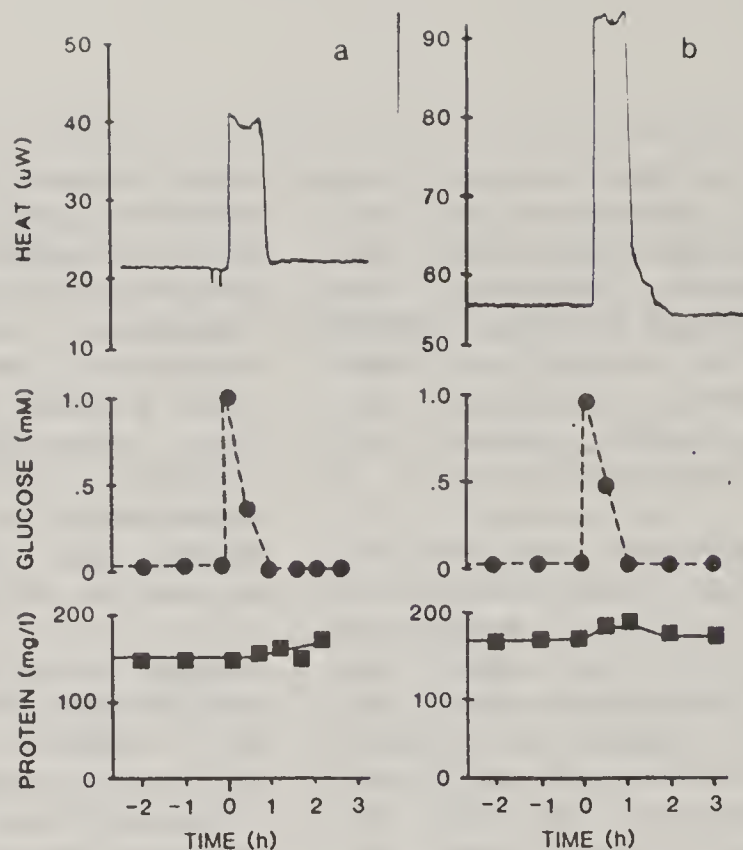[1] Equal parts starch, sucrose, cellobiose, xylan and pectin.

Figure 1. Effect of pulse dose of glucose (1 mM at time 0) on the heat production of S. ruminantium (a) and B. ruminicola (b).

CONCENTRATION GRADIENT OF AMMONIA ACROSS THE CELL MEMBRANE OF MIXED RUMEN BACTERIA

J.B. RUSSELL AND H.J. STROBEL

INTRODUCTION

Ammonia is an important nitrogen source for the growth of rumen bacteria in vivo, but excessive ammonia in the rumen can be deleterious to the animal. Ammonia is a lipophlic substance that can be absorbed across the rumen wall into blood. The animal detoxifies ammonia by synthesizing urea, but urea synthesis is costly. Thus, the urinary excretion of urea represents a loss of both nitrogen and energy. A deficiency of ammonia in the rumen, however, can decrease the efficiency of microbial protein synthesis. There has been considerable disagreement concerning the optimal concentration of ruminal ammonia (Satter and Slyter, 1974; Mehrez et al., 1977; Schaefer et al., 1980). Little was known about the mechanism of ammonia transport in rumen bacteria. Active transport mechanisms would allow them to grow rapidly, even if ammonia concentrations were low. If ammonia entered the cells by passive diffusion, energy would not be needed for uptake, but higher concentrations would be needed. Since the establishment of a concentration gradient is the most basic

characteristic of active transport, we decided to measure the concentration gradient of ammonia across the cell membranes of rumen bacteria.

MATERIALS AND METHODS

Mixed rumen bacteria were grown in carbohydrate limited batch cultures and the concentration of ammonia in the incubation medium was varied from 0 to 500 mg/l. Intracellular volume was estimated from the difference between ^{14}C-polyethylene glycol and $^{3}H_2O$ counts in cells that were centrifuged through silicon oil. Intracellular and extracellular ammonia were measured with a colorimetric assay (Chaney and Marbach, 1962). Carbohydrate utilization was measured by the phenol-sulfuric acid assay, protein was measured by the Lowry (1951) assay, and fermentation acids were quantified by high pressure liquid chromatography.

RESULTS

When mixed ruminal bacteria were provided with growth rate limiting amounts of mixed carbohydrates, more than 50 mg ammonia per liter were required for maximal protein synthesis. If ammonia concentration was less than 50 mg per liter, microbial protein synthesis declined and there was an increase in unfermented carbohydrates. Ammonia starvation also resulted in decreased growth efficiency. Intracellular ammonia increased as a linear function of extracellular ammonia, but the intracellular concentration was always more than 160 mg per liter higher than the extracellular concentration. Maximal protein synthesis was not observed until intracellular ammonia was greater than 220 mg per liter. The concentration gradient of ammonia across the cell membranes

ranged from 1.5 to 1.8 fold and indicated that some of the ruminal bacteria may have active transport mechanisms for ammonia. These concentration gradients were, however, far less than values reported for bacteria from other habitats. The ruminal bacteria left more than 12 mg ammonia per liter when carbohydrates were still available, and this observation was consistent with the assumption that active ammonia transport was not maximally or readily induced.

DISCUSSION

Active transport systems for ammonia in non-rumen bacteria are induced by ammonia and repressed by high levels of ammonia. When bacteria are grown in nitrogen limited chemostats (or transferred for long periods in nitrogen limited batch cultures), there is intense selection for ammonia assimilation, the physiological characteristic limiting growth. Under these conditions active transport systems for ammonia could be induced. In vivo the selection pressure on ruminal bacteria is not constant. Poor quality diets are usually limiting in energy, as well as nitrogen, and even nitrogen availability is inconsistent during the feeding cycle. Since active transport of ammonia would be driven by ATP hydrolysis and a resulting protonmotive force, the induction of an ammonium carrier would be at the expense of energy utilization.

Our results agreed with those of Satter and Slyter (1974), but the observed requirement for ammonia (50 mg per liter) was significantly less than the value reported by Mehrez et al. (1977). This difference may be related to the bacterial growth rates. In Satter and Slyter's and our experiments,

bacterial growth rates were less than 0.09 h^{-1} while Mehrez et al. used the maximal fermentation rate of barley as their criterion. Growth rates were not reported in the latter set of experiments and estimations of growth rate were confounded by lag time. Barley starch, however, often ferments at a rate greater than 0.30 h^{-1}. In accordance with the basic premise of affinity and Michaelis Menten kinetics, higher concentrations of the limiting substrate (ammonia) should be required for greater velocity (growth rate) and the data of Erdman et al. also support this concept.

A PROPOSED MECHANISM OF MONENSIN ACTION IN INHIBITING RUMINAL BACTERIAL GROWTH: EFFECTS ON ION FLUX AND PROTONMOTIVE FORCE

J.B. RUSSELL

INTRODUCTION

Methane production varies with the type of diet fed, but it usually accounts for 5 to 10% of the energy in the diet and is a significant loss to ruminants. Ionophores like monensin kill ruminal bacteria that produce hydrogen, a precursor of methane, and select for bacteria that produce either succinate or propionate. In spite of widespread application, little was known about the biochemical mechanisms of these compounds in ruminal bacteria. Streptococcus bovis is a gram-positive anaerobe that proliferates in the rumen when large amounts of grain are fed, and it is sensitive to monensin. The beneficial responses of this drug may be at least in part related to inhibition of this organism.

MATERIALS AND METHODS

S. bovis JB1 was grown in medium containing salts, cysteine hydrochloride, yeast extract, Trypticase, and 20 mmol glucose per liter. Intracellular pH was measured by an acid distribution method that was originally described by Riebeling et al. (1975). This method is based on the assumption that nondissociated forms of weak acids diffuse through the lipophilic cell membrane freely so that the internal and external concentrations equilibrate. Distribution of the ionized form of the acid then becomes a function of the gradient between internal and external pH. Intracellular space was calculated from the difference in specific activity of ^3H_2O and [1,2 ^{14}C]-polyethylene glycol. Intracellular pH was calculated from the specific activity of [7 ^{14}C] benzoate in the supernatant and the pellet using the Henderson-Hasselbach equation. The chemical potential gradient (ZΔpH) generated by the pH gradient across the cell membrane was calculated from the Nerst relationship. The electrical potential ($\Delta\Psi$) generated by the pH gradient and the subsequent movement of the ions was calculated from the uptake of [phenyl ^{14}C]

tetraphenylphosphonium bromide. The total transmembrane potential (Δp) was calculated from $p = \Delta \Psi - Z \Delta pH$.

RESULTS

S. bovis was unable to grow in the presence of monensin. When monensin (5 mg/liter) was added to actively growing cultures, there was an immediate decrease in growth rate and within 3 h no further growth was observed. Glucose utilization and lactate production, however, continued for another 8 h even though growth had ceased. Monensin caused a decrease ($P<.05$) in intracellular K^+, a decrease ($P<.05$) in intracellular pH, and an increase ($P<.05$) in intracellular Na^+. The net exchange of K^+ for Na^+ and H^+ via monensin was driven by the difference in concentration of K^+ and Na^+ across the cell membrane. Nontreated cells maintained a 70-fold gradient (inside higher) for K^+ while the Na^+ gradient was only 2.7 fold (Table 1).

DISCUSSION

Monensin is usually described as an antiporter that facilitates a one-for-one exchange of H^+ and Na^+ across cell membranes; however, it can also mediate an H^+ and K^+ exchange (Pressman and Fahim, 1982). Since the K^+ gradient was much greater than the Na^+ gradient, K^+ efflux via monensin was more exergonic than Na^+. K^+ in turn lead to an accumulation of H^+ and a decrease in intracellular pH. Once the intracellular pH was lower inside than outside influx of Na^+ could be driven by H^+ efflux. Growth inhibition probably resulted from the utilization of ATP to expel an excess of H^+ from the cell. Dawson and Boling (1983) reported that high concentrations of K^+ decreased the sensitivity of ruminal bacterial to monensin, our model explains the effect of added K^+. Previous models had proposed an opposite effect whereby Na^+ would leave, not enter, the bacteria.

Table 1. Effect of monensin on the membrane potential of S. bovis.

Measurement[a]	Control	Monensin
pH_e	$6.65 \pm .02$[b]	$6.65 \pm .02$
pH_i	$7.08 \pm .15$	$6.20 \pm .13$[c]
$Z\Delta pH$, mV	-26 ± 9.2	28 ± 8.3[c]
$\Delta \Psi$, mV	77 ± 6.9	71 ± 3.9
Δp, mV	103 ± 2.3	43 ± 4.4[c]
V_i, ul/mg protein	$6.0 \pm .5$	$5.1 \pm .11$
K_e, mM	$9 \pm .3$	$9 \pm .5$
K_i mM	613 ± 13.4	134 ± 11.1[c]
Na_e, mM	89 ± 4.7	93 ± 6.9
Na_i, mM	237 ± 47.4	543 ± 7.1[c]

[a] "e" indicates extracellular and "i" indicates intracellular.
[b] Student's test (n=4).
[c] Means within a row which differ ($P<.05$) from the control.

NUTRITIVE QUALITY OF COOL- AND WARM-SEASON GRASSES GROWN IN DIFFERENT IRRADIANCE REGIMES

K.D. KEPHART AND D.R. BUXTON

INTRODUCTION

Irradiance (light) is one of the three major cardinal environmental factors that influence plant growth. Adaptation to irradiance regimes directly influences morphological characteristics such as specific leaf weight and leaf-to-stem ratio, as well as crop growth rate and yield. Forages, perhaps to a greater degree than other crops, are grown under a variety of irradiance conditions. High plant densities or mixtures of tall and short species lead to shading within the canopy. At the temperate latitudes, forages are grown under a range of photoperiods consisting of short days during spring and fall, and long days during the summer. Furthermore, forages are shaded during periods of cloudiness. Cool- and warm-season grasses may differ in nutritive quality response to irradiance because of known differences between cool- and warm-season grasses in photosynthetic response to irradiance regimes. The objective of this investigation was to compare cool- and warm-season grasses for response of nutritive quality to growth under reduced irradiance.

MATERIALS AND METHODS

Two warm-season grasses (big bluestem and switchgrass) and three cool-season grasses (deertongue, reed canarygrass, and tall fescue) were grown under three levels of irradiance; 37, 70, and 100% of available irradiance. The leaves, stems, and total herbage were sampled on three occasions during spring growth of 2 years. The plant material was analyzed for in vitro digestible dry matter (IVDDM), neutral-detergent fiber (NDF), acid-detergent fiber, acid-detergent lignin, and total nitrogen.

RESULTS AND DISCUSSION

The IVDDM concentration of stems, leaves, and total herbage generally decreased linearly with increasing irradiance. The average IVDDM in herbage of plants grown under 37% available irradiance was 56.4%, compared to an average IVDDM of 53.7% in herbage of plants grown under 100% available irradiance. The decrease in IVDDM with increasing irradiance was associated with increased NDF concentration and decreased nitrogen concentration. Lignin expressed on a cell-wall (NDF) basis generally increased with increasing irradiance. The response of the cool- and warm-season grasses was similar. The only clear difference between the cool- and warm-season grasses was in NDF concentration in leaf tissue and total herbage. The average NDF concentration in leaf tissue of the warm-season grasses was 18% greater than in the cool-season grasses. The results suggest that shading during plant growth caused a photosynthetic limitation to cell-wall development which resulted in higher forage digestibility.

CROP WATER STRESS INDEX AND YIELD OF
WATER-DEFICIT-STRESSED ALFALFA DURING GROWTH

M.J. HATTENDORF, R.E. CARLSON, R.A. HALIM, AND D.R. BUXTON

INTRODUCTION

Remote sensing technology has been developed in an effort to use satellite sensors to assess crop yields. Hand-held infrared themometry is an application of this technology that has proven potentially useful in irrigation scheduling and yield prediction of water-deficit-stressed crops because water-stressed plants have a higher canopy temperature than nonstressed plants. Canopy-temperature indices such as canopy minus air temperature (Tc-Ta) and the Crop Water Stress Index (CWSI) have been used to assess relationships of alfalfa yields under water stress, but at only one time during the alfalfa growth cycle. Alfalfa and other perennial forage crops are normally harvested while immature; the stage depends upon factors such as desired forage quality and the prevailing weather. Because alfalfa continues to grow and increase in yield during the growth cycle, there is no "finite" forage yield. The possibility exists that the alfalfa yield:CWSI relationship is dynamic and that yield prediction of water-stressed alfalfa might have to be adjusted for the length of growth. The objectives of this study were to define the alfalfa yield:stress index relationship, to test the possible advantages of using the CWSI over the Tc-Ta differential, to apply the double-exponential Gompertz growth function to yields over time, and to develop a response surface encompassing the Gompertz model and the yield:CWSI relationship.

MATERIALS AND METHODS

'Apollo II' alfalfa was grown in 100 L containers set into the ground and protected by a movable rain-out shelter. Plants were watered either weekly or twice weekly to 112, 100, 88, 77, and 65% of field capacity during 2 years. Regrowth herbage was harvested at 5 weekly intervals beginning 3 weeks after the initial cut. Canopy temperature was measured daily with a hand-held infrared thermometer. Wet and dry bulb temperatures for determination of vapor pressure deficit were collected concurrently between 1200 and 1400 h using an aspirated psychrometer. These data were used to calculate daily CWSI and Tc-Ta. Water use was determined by the soil-water balance technique.

RESULTS AND DISCUSSION

The water use:yield and CWSI values for the 2 years of study indicated that the data could be combined into a single response surface even though yields were higher in the 2nd year than in the 1st year. The alfalfa yield:CWSI relationships were exponential curves that became increasingly curvilinear with later harvests. Thus, the alfalfa yield:CWSI is a dynamic process and is not adequately described by a single curve. The Tc-Ta:yield relationships lost the curvilinearity that was characteristic of the yield:CWSI relationship, but the 2 years of data did not fit the same line. The

Tc-Ta index seems to have little utility in yield:stress comparisons between years. The Gompertz function adequately modelled the water-stressed alfalfa yields except when sudden stress caused leaf drop. Values of coefficients of determination ranged from 0.74 to 0.98. The exponential yield:CWSI relationship was combined with the Gompertz function for yield:time relationships for development of a response surface. The response surface equation had an approximate coefficient of determination of 0.65 with n=250. Gradients of water-stressed alfalfa yields were better described by a family of exponential curves than by a single relationship. We conclude that the CWSI is a more appropriate stress index for comparison or use with more than 1 year of data at the same location than the Tc-Ta stress index and that the Gompertz equation is useful in describing yields of water-stressed alfalfa over time, provided that the alfalfa has not suffered biomass loss.

PECTIC POLYSACCHARIDES FROM ALFALFA

R.D. HATFIELD

INTRODUCTION

The pectic fraction of plant cell walls is composed primarily of polymers containing polygalacturonic acid, but does contain polysaccharides composed of neutral sugar residues. This fraction is normally found in the primary cell wall and is heavily deposited in the region known as the middle lamella. The middle lamella also contains lignin and may be the site of initial lignification. Although pectic materials are thought to be completely degraded in the rumen, the rate of degradation could influence the rate of digestion of other cell wall components. Interactions with lignin or other cell wall components could alter degradation patterns of the pectic fraction. The objective of this study is to characterize the type of polysaccharides and to identify molecular interactions within the pectic fraction of the alfalfa cell-wall matrix.

MATERIALS AND METHODS

Alfalfa (Medicago sativa) plants were grown in a greenhouse and harvested at early flower-bud initiation. Plants were freeze-dried and the stems were analyzed; each was separated into lower, middle, and upper zones. Cell walls were isolated from the lower zone and subjected to sequential extractions. The extraction sequence consisted of H_2O (75-80°C, 1h), 0.5% $(NH_4)_2C_2O_4$ (70-80°C, 1h), 1.0% $(NH_4)_2C_2O_4$ (70-80°C, 1h), acidic $NaClO_2$ to remove lignin and a repeat of the 1.0% $(NH_4)_2C_2O_4$ extraction. Each fraction was dialyzed 20 h against distilled H_2O and freeze-dried. The recovered extracts were weighed and analyzed for total uronic acids and neutral sugar composition as alditol acetates by using gas chromatography. The samples of the $(NH_4)_2C_2O_4$ extraction and the lignin extraction were

resuspended in 20 mM sodium acetate buffer (pH 5.0, 20 mM NaCl) and subjected to DEAE ion exchange chromatography. Bound carbohydrate was eluted with NaCl gradient (20 mM to 500 mM). Collected fractions were analyzed for uronic acid and total sugar content. Pooled peak fractions were analyzed for neutral sugar composition as alditol acetates.

RESULTS AND DISCUSSION

The five extractions solubilized 20% of the total cell wall matrix. Uronic acid composition ranged from 33% in the lignin extract to 68% in the 0.5% $(NH_4)_2C_2O_4$ fraction (Table 1). The molar ratios of rhamnose, fucose, mannose, and galactose were similar while xylose and arabinose showed greater variability between fractions (Figure 1). This indicates that the neutral sugar components of the pectic fractions are different and represent different branching patterns or changes in associated polysaccharides. A comparison of the DEAE column profiles of the 0.5% $(NH_4)_2C_2O_4$ extract with the lignin extract indicates that the major portion of the carbohydrate for both fractions was bound to the column. The unbound, neutral fraction represented about 10-20% of the total carbohydrate. Neutral sugar composition of the unbound fractions was similar, being composed of arabinose, xylose, galactose and glucose. This suggests that the neutral polysaccharides solubilized by these treatments are most likely arabinans, arabinogalactans, galactans and/or xyloglucans. Although the bound polysaccharides eluted as a single broad peak, the ratio of total sugar to uronic acid content in the fractions indicated a heterogenous mixture of acidic polysaccharides. Neutral sugar composition of the more tightly bound fractions contained higher molar ratios of rhamnose, suggesting less branched polysaccharides.

These preliminary findings demonstrate that the pectic portion of alfalfa stem-cell-wall matrices contains a heterogenous mixture of polysaccharides. Lignin extraction solubilizes both neutral and acidic polysaccharides with similar compositions to typical pectic materials. This denotes a close interplay between lignin and specific pectic-fraction polysaccharides through hydrogen, ionic or covalent bonding. Such an interaction is supported by the observation of increased solubilization of pectic material after lignin removal (Table 1).

Table 1. Cell Wall Extractions

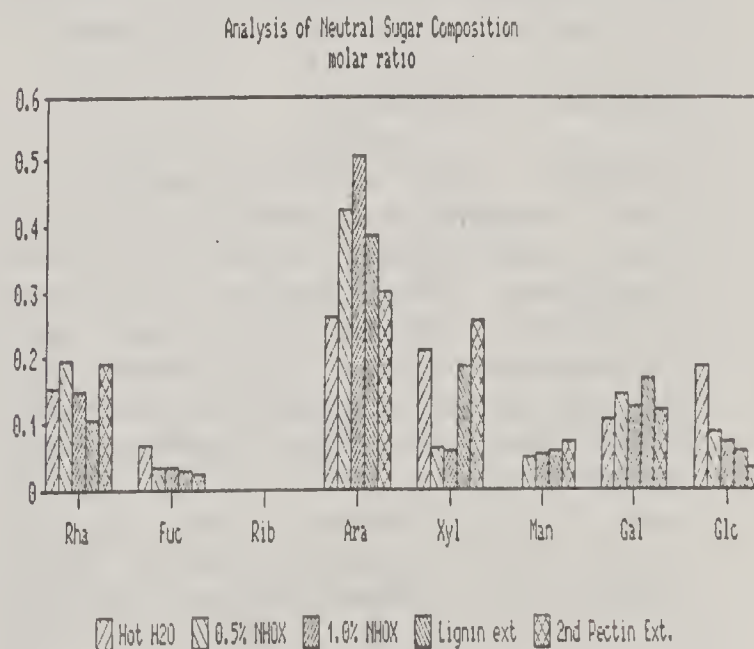| Extraction	Recovery mg/g CWM	% Uronic Acid
hot H_2O	5.0	36.8
0.5% oxalate	38.2	67.9
1.0% oxalate	21.7	52.7
lignin ext.	93.6	33.0
1.0% oxalate	41.8	60.8

Figure 1

RECURRENT SELECTION FOR 2N POLLEN FORMATION IN RED CLOVER

R.R. SMITH AND W.A. PARROTT

INTRODUCTION

The discovery of 2n gametes (gametes with the chromosome number of the sporophyte) in red clover, Trifolium pratense L., and the ability to produce polyploids in this species indicate their possible use to routinely develop tetraploid ($2n = 4x = 28$) red clover. Red clover occurs naturally as a diploid ($2n = 2x = 14$) species, but tetraploid strains developed in Europe frequently out-perform diploid strains suggesting it would be desirable to develop tetraploid germplasm adapted to the red clover-growing regions of the USA. A high frequency of 2n pollen production is a key to successful sexual tetra-ploidization in red clover, such that tetraploid seed would be obtained following either 4x-2x or 2x-2x crosses. Recurrent selection is one method to develop a population with a high frequency of 2n pollen formation. Therefore, a recurrent selection program was initiated in an attempt to develop a red clover population with a high frequency of 2n pollen formation.

MATERIAL AND METHODS

Eighteen red clover plants (C_0) that produced at least 1% 2n pollen were identified initially after screening 600 plants from six cultivars. These C_0 plants were intercrossed using honey bees (Apis mellifera L.) to effect pollination. Progenies from these matings were designated as the C_1 population and 25 C_1 progeny from each C_0 plant were evaluated for 2n pollen production. The highest fifteen C_1 2n pollen producers were selected and intercrossed to produce the C_2 generation. Again 25 C_2 progeny from each selected C_1 plant were evaluated for 2n pollen production and the highest thirty C_2 2n pollen producers were selected and intercrossed to produce the C_3 generation.

To measure progress from selection for 2n pollen production, 60 plants representing each cycle were evaluated in the greenhouse in a replicated complete block test. Narrow sense heritability (h^2) was estimated as a ratio of the additive genetic variance to the phenotypic variance. Realized heritability was calculated for each cycle by dividing the realized gain by the selection differential.

RESULTS AND DISCUSSION

Average frequency of 2n pollen production per plant increased from 0.04% in the base cycle (C_0) to 47.38% in the third cycle (Table 1). There were highly significant differences among cycles with the greatest increase realized between cycle 2 and cycle 3. Both the frequency of 2n pollen production per plant and the frequency of plants that produce 2n pollen in each cycle increased. Seven, 75, 97, and 100% of the plants produced 2n pollen in the C_0, C_1, C_2, and C_3 cycle, respectively.

Significant differences were observed between cycles for dry weight. A slight increase was observed between the C_0 cycle and C_1 which was undoubtedly due to heterosis. This difference appeared to persist since dry weight per plant in cycle 3 was greater than the base cycle, C_0.

Recurrent selection was effective in increasing the frequency of red clover plants which produce 2n pollen. It would appear that the derived population, C_3, would serve as a broad-based, heterozygous population that will lead eventually to the development of a tetraploid red clover.

Table 1. Mean percent 2n pollen production and dry weight (g) per plant for three cycles of selection for 2n pollen production in red clover.

| | 2n pollen | | Dry |
Cycle	Mean	Range	Weight
0	0.04	0.0-1.20	4.09
1	3.09	0.0-59.5	5.02
2	17.86	0.0-88.3	4.26
3	47.38	0.8-99.6	4.71

PERFORMANCE OF THE NEW GENERATION OF RED CLOVER CULTIVARS

R.R. SMITH AND D.K. SHARPEE

INTRODUCTION

One of the reported disadvantages of red clover as a forage is its lack of persistence or ability to be productive for more than one harvest year. This poor persistence has been attributed to the lack of winterhardiness and to disease susceptibility. Recent improvements in red clover germplasm relative to disease resistance and yield should contribute to greater longevity of this species. This study was designed to ascertain the impact of the new generation of red clover cultivars.

MATERIAL AND METHODS

Three red clover cultivars, (Arlington, Redman, and Prosper I) and two Wisconsin Experimentals (WHC28 and WHC30) developed after 1970 and five cultivars (Pennscott, Chesapeake, Sterling, Lakeland, and Common) developed prior to 1970 were established on the Arlington Experimental Station in a randomized complete block design with four replications in 1984. Three harvests were taken in each of two years, 1985 and 1986. Reaction to the northern anthracnose disease was determined prior to the first harvest and percent ground cover was recorded as percent stand in April and October of the second harvest year, 1986.

RESULTS AND DISCUSSION

Red clover cultivars developed and released after 1970 (new) were

superior in dry matter yield than those developed prior to 1970 (old) (Table 1). New cultivars exceeded the old cultivars by 50% in the second harvest year (3.64 vs. 2.01 tons DM/A). Assuming $11.00 per cwt for milk, the new cultivars would average $412 greater return per acre per year than the old cultivars. This translates to $824 improvement in dollar return per acre over the two harvest years. After two harvest years the new cultivars average 60% stand in contrast to only 10% for the old cultivars suggesting a substantial improvement in persistence. The new cultivars are more resistant to northern anthracnose. The improvement in the performance of the newer cultivars is attributed to improved persistence and disease resistance (through selection) and to improved yield per se. The large difference (1.6 tons) between the two groups in the second harvest year was undoubtedly underestimated since the yield of poor stands is often made up of weedy grasses and broadleaf plants. Forage stands of less than 25% would normally be destroyed and new seedings established. This would have been the recommendation if old cultivars were used; however, a stand of 62% for the new cultivars, which yielded 3.7 tons of dry matter the second year, would suggest the retention of the stand for an additional year.

While new cultivars of red clover do not persist as well as alfalfa in fertile, well drained soil, recent improvements in the germplasm would suggest that the new cultivars do persist through the second harvest year. Red clover is well adapted to poorly drained, more acid soils. Such conditions exist in many areas in the northcentral states. The new cultivars of red clover are well adapted to these areas.

Table 1. Mean dry matter yield (TDM/A), percent stand, northern anthracnose (NA) and milk yield (milk/A) for red clover cultivars developed prior to and after 1970 grown at Arlington, WI in 1985 and 1986.

Cultivars Developed	Yield			Milk/a[+]	Stand		NA[++]
	1985	1986	85-86		4-86	10-86	
	(TDM/A)			(lb/a)	(%)	(%)	
After 1970 (NEW)	5.36	3.64	9.01	11,100	65	60	2.4
Before 1970 (OLD)	4.00	2.01	6.01	7,400	27	10	4.4
Difference (%)	38	81	50	50	241	600	--

[+] Calculations based on Wisconsin Milk Production formula; assume 1350 lb. animal; 80 lb. milk/day with 4.0% fat; red clover with 20% crude protein, 26% ADF and 39% NDF.

[++] Scale: 1 = resistant, no symptoms; 5 = susceptible, 90% of plants with symptoms.

STRUCTURAL NEUTRAL SUGARS IN LEGUME AND GRASS STEMS IN RELATION TO DIGESTIBILITY

D.R. BUXTON, J.R. RUSSELL AND W.F. WEDIN

INTRODUCTION

Forage digestibility by dairy cows and other ruminants is limited primarily by the amount of cell wall in herbage and its degradability. Cell content, the complement of cell wall, is nearly completely digestible. Cell walls, on the other hand, have lower digestibility, depending upon their chemical and physical structure. Cell walls are comprised primarily of cellulose microfibrils and the hemicellulose and lignin matrix in which they are embedded. Cell-wall digestibility is faster in legumes but more complete in grasses. Lignin is the chemical most responsible for limiting the extent of cell-wall digestion, but the composition and structure of polysaccharides in the cell-wall matrix can exert additional influence in some situations.

Components of cell walls frequently are determined by gravimetric methods, such as the detergent-fractionation system. These methods sometimes fail to accurately predict digestibility for a range of forages grown in different environments. Some recent studies have shown that separation of cell-wall polysaccharides into their constituent monosaccharides helps to explain differences in digestibility among forage species and plant parts. We conducted this study to determine the concentration of neutral sugars from polysaccharides in cell walls of stems of immature and mature grasses and legumes and to relate neutral sugar concentrations to holocellulose concentration and to in vitro digestibility of dry matter.

MATERIALS AND METHODS

In vitro true digestibility (IVTD); neutral-detergent fiber, acid-detergent fiber, permanganate lignin, and acid-detergent insoluble ash (used to estimate holocellulose concentrations); and neutral sugars in ethanol-insoluble residues were determined in 15 cm segments of stem basis of field-grown cultivars of alfalfa, birdsfoot trefoil, red clover, smooth bromegrass, and orchardgrass.

RESULTS AND DISCUSSION

The concentration of total neutral sugars, converted to polysaccharide equivalents, averaged 90% of holocellulose values in the legume and mature grass stems and 81% of holocellulose values in immature grass stems. In immature stems, the concentration of total neutral sugars was greater in grasses than in legumes, whereas in mature stems, the concentration of total neutral sugars did not differ greatly between grasses and legumes.

The ratio of hemicellulose:lignin in the cell-wall matrix ranged from up to ten in immature grass stems to as low as three in mature grass stems. Conversely, the ratio in the legumes was near one and was not influenced by maturity. Glucose comprised 62% of the total neutral sugars in the grasses and 67% in the legumes; xylose comprised 30% in grasses and 20% in legumes. The proportion of glucose increased and the proportion of arabinose decreased with maturity. The arabinose proportion was similar for grasses and legumes

whereas galactose, mannose, and rhamnose were present in larger amounts in the legumes than in the grasses. Differences in proportions of these neutral sugars also were found within the grasses and within the legumes.

Among the grasses and among the legume species, IVTD concentration was closely related to lignin concentration and, in spite of large differences in proportions of these neutral sugars, the only sugar that was associated with IVTD independent of lignin was mannose in grass stems. Thus, we found important differences in chemical composition in stems of grass and legume polysaccharides, but lignin exerted the dominant control over the extent of digestion with very little additional effect that could be attributed to differences in polysaccharides.

CHANGES IN MINERAL COMPOSITION OF FOUR TROPICAL PASTURE FORAGES DURING THE WET SEASON IN MALAWI, AFRICA

F.A. MARTZ, J.F. OLBRICH, M.S. KUMWENDA, AND R.L. BELYEA

INTRODUCTION

Malawi, a third world nation, is located in South Central Africa. It is a landlocked country with a population of 5.6 million. The climate is characterized by constantly high temperatures and high humidity. Rainfall is often of the convectional type. Annual precipitation often reaches 1.5 m, but may fall below 0.8 m in dry areas. In many areas the year is divided into a wet season (January-May) and a dry season (June-December).

Malawi is predominantly agricultural and livestock is an important enterprise. Both dairy and beef production are practiced and the dairy production is regionalized into three milkshed areas around major cities, Blantyre, Lilongiwe and Mzuzur.

Productivity of livestock is low throughout the country. Extensive losses occur from disease and malnutrition and levels of production are among the lowest found anywhere. These livestock are nearly entirely dependent on natural grasslands. Pastures are usually low in quality and high in moisture content.

Little is known about the effect of forage species and season on the nutrient composition of pasture forages in Malawi. Accurate supplementation is impossible without this information.

OBJECTIVES

The objective of this study was to determine the nutrient content as well as changes in nutrient content of five pasture forages which were grown during the wet season (January-May).

MATERIALS AND METHDOS

The samples analyzed in this study originated from a project conducted by the University of Florida/USAID/ Malawi Research (UF/USAID/MA Project). This project was conducted at a research station at Malawi University-Lilongiwe near Lilongiwe, in Central Malawi.

There were five treatments: (1) Star Grass (ST), (2) Giant Panic (GT), (3) Rhodes Grass (RG), (4) Rhodes Grass (cv Mbara) plus Silverleaf (desmodium) (RGMS), and (5) Rhodes Grass (cv Boma) plus Silverleaf (RGBS). Fifteen pasture plots (1.5 Ha) were layed out, three replicates per treatment. Each plot was divided into four subplots for sampling purposes. These pastures were seeded in November-December 1983 and pastures were sampled January through May 1984.

Samples were collected approximately each week for the 5-month period. Pastures were grazed rotationally using Malawi-Zebu steers which were moved to a new pasture each week. Thus, pastures had a 21 d rest period which resulted in full regrowth of the pastures.

Samples were taken immediately prior to turning cattle into the new pasture. Samples were collected by hand clipping a 2 m area at a stubble height of 15 cm. To form the final sample set, replicate samples were combined for each treatment for each week to make 100 samples (20 weeks x 5 treatments).

Samples were analyzed for fiber, protein and in vitro digestibility by the Van Soest procedures. Calcium, Phosphorus, Potassium, Magnesium, Iron, Manganese, Sodium, Zinc and Copper were analyzed by atomic absorption. Standard forage samples as well as National Bureau of Standard samples were analyzed to insure analytical accuracy.

RESULTS AND DISCUSSION

Data for fiber and protein composition and in vitro digestibility are shown in Table 1. Even in a wet tropical climate, changes over season in composition were not unlike the changes seen for warm season grasses in temperate climates. Fiber content increased and protein and dry matter digestibility decreased as the season progressed. Protein declined especially rapidly and by April and May was at or below critical levels.

The addition of Silverleaf, a legume, to the pastures would be expected to improve nutrient composition. These changes appeared to occur with Rhodes Grass (Boma) plus Silverleaf where protein was increased and NDF and ADF decreased compared to the other pastures. The Rhodes Grass (Mbara) plus Silverleaf pastures did not appear to benefit nearly as much from the Silverleaf. This lack of improvement may be due to a poor stand of Silverleaf in these pastures. We do not have data for the botanical composition of the pastures.

The mineral composition for the pastures is shown in Table 2. Differences among forages (P<.05) were found in several minerals. Calcium, Potassium, Iron and Manganese levels were adequate for both growth and lactation. Magnesium, Sodium, Phosphorus, Zinc and Copper were either borderline or deficient for growth and lactation. Most mineral levels declined during season. Silverleaf (legume) did not improve mineral levels significantly. Data indicate potential for supplementation of Malawi pastures with P, Mg, Na, Zn, and Cu.

Table 1. Chemical composition and in vitro digestibility of five pasture
          forages grown at Lilongiwe, Malawi.

Forage Species	Month	Average Composition for Month			
		NDF	ADF	Protein %	IVDMD
ST	January	72	32	17	80
	February	74	36	13	68
	March	77	41	10	61
	April	74	41	9	57
	May	79	43	8	53
GP	January	69	35	15	82
	February	69	38	12	74
	March	69	38	12	75
	April	72	42	10	73
	May	67	41	9	71
RGMS	January	66	35	15	78
	February	71	39	13	69
	March	77	42	9	65
	April	75	45	8	59
	May	76	44	7	57
RGBS	January	48	32	20	71
	February	51	36	21	63
	March	54	38	19	60
	April	52	41	16	53
	May	54	43	12	55
RG	January	71	37	14	79
	February	74	39	12	72
	March	72	39	12	70
	April	76	46	7	59
	May	77	47	6	52

Table 2. Mineral composition of five pasture forages grown at Lilongiwe, Malawi.

Forage Species	Month	Ca	P	K	Mg	Na	Fe	Mn	Cu	Zn
						Mineral				
		%------------					----------ppm----------			
ST	January	.65	.03	3.0	.19	.03	344	89	13	26
	February	.58	.02	2.5	.17	.05	360	93	12	23
	March	.54	.02	2.4	.13	.05	267	87	11	21
	April	.45	.02	2.3	.14	.04	226	80	11	19
	May	.36	.01	2.0	.14	.03	207	67	11	22
GP	January	.61	.03	3.4	.07	.02	268	123	9	16
	February	.63	.02	2.9	.07	.03	262	130	8	19
	March	.60	.02	3.0	.07	.02	208	114	8	19
	April	.51	.02	2.9	.06	.02	135	102	7	19
	May	.47	.01	2.4	.08	.02	100	87	6	20
RGMS	January	.76	.03	6.5	.16	.02	379	97	9	17
	February	.64	.03	5.8	.17	.03	344	68	8	15
	March	.53	.02	5.8	.14	.02	295	62	7	15
	April	.47	.02	4.6	.13	.03	221	58	7	13
	May	.39	.02	3.9	.14	.02	187	53	6	13
RGBS	January	.73	.03	6.3	.16	.02	378	93	7	19
	February	.64	.03	5.8	.17	.03	343	68	8	20
	March	.53	.02	5.6	.14	.02	297	61	8	14
	April	.46	.02	4.6	.13	.03	221	58	7	12
	May	.39	.02	3.5	.13	.02	186	51	6	14
RG	January	.69	.03	6.0	.16	.03	322	69	8	19
	February	.66	.03	5.5	.17	.03	269	65	8	17
	March	.60	.02	5.7	.15	.03	230	68	7	17
	April	.57	.01	4.3	.11	.02	201	57	7	14
	May	.40	.01	2.8	.10	.02	182	47	6	15

LEAF LOSS IN FORAGE HARVEST USING
DRYING AGENTS AND PRESERVATIVES

R.P. WALGENBACH

INTRODUCTION

There is considerable interest in using chemical conditioning and preserving agents to reduce field curing time and dry matter losses in baling and storing of forage crops. At last year's annual meeting I presented data on the efficacy of a chemical conditioning agent and of several preserving agents. The data indicated that chemical conditioning agents can significantly reduce field curing time under good drying conditions; however, inconsistent curing occurs under poor drying conditions. Of several preserving agents tested, only one containing a large proportion of propionic acid (80% propionic and 20% acetic) gave any indication of good preservation. Many claims are made for the potential dry matter savings to be had by using preserving agents. Our objectives in this research were to quantify baling and storage losses when chemical conditioning and preserving agents are used.

METHODS

The chemical conditioning agent containing approximately 50% sodium carbonate and 50% potassium carbonate was applied using 30 gals. of water per acre. The preserving agent was applied in a 50% solution with water using nozzles mounted to spray the top and the bottom of the windrows. At application rates of 30 lbs/ton, no water was used. The preserving agent contained a mixture of 80% propionic and 20% acetic acids. The baling times and application rates of chemical conditioning and preserving agents are provided in table 1. Baling losses were collected by sheathing a John Deere 336 baler with a pre-formed polyethylene tarp. This tarp was designed to minimize losses due to wind. The losses occurring at the baler pick-up mechanisms were estimated from data obtained in other studies. Storage losses were determined by obtaining initial and after storage bale weights and moisture contents.

RESULTS AND DISCUSSION

The losses as a percent of potentially harvested dry matter are presented in Table 1. Losses ranged from 3.6 to 9.4% for hay that was not rained on. In Study 2 hay was left to be rained on and this loss was near 40%. In Study 1 mean moisture contents ranged from 16 to 24%; however, keep in mind that actual bale moisture ranges were much greater. The driest hay in these experiments (13%) occurred with chemically conditioned alfalfa in Study 2. Excellent drying conditions occurred during this study and a complete set of treatments was not obtained.

The wettest hay was baled in Study 3 which contained 32% moisture at baling. The total baling loss difference between the driest hay (5.0%) and that of the wettest hay (2.3%) was 2.7%. This difference, while significant, is much lower than one might expect. Claims have been made that chemically conditioned alfalfa retains its leaves better than that not chemically conditioned. But, there is little data available to support such an observation. The least amount of DM was lost during baling when previously dried alfalfa was

rewetted with evening dew and baled after 9:00 p.m. or in early morning hours with a preserving agent.

The highest storage losses occurred with the wettest hay in Study 3. In general, wet hay treated with the 80P:20A acid mixture had relatively low dry matter losses during storage. This hay was stored in somewhat small stacks which probably influenced dry matter losses.

Whether or not to use chemical conditioning agents or preservatives is not a simple or clear cut decision. Factors such as weather risks are very important, saving high quality dry matter such as leaves is important but may not always be economic, and time management is another consideration. Give careful thought to such factors in your operation needs.

Table 1. Influence of a chemical conditioning agent (CCA), a preserving agent (PA), bale moisture content, and baling time on alfalfa harvest and storage losses.

| Treatment | Time | Mean Bale Moisture Content | Bale Loss | | Storage Loss | Total Loss |
			At[1] Pick-Up	After[2] Pick-Up		
			----------------------------%----------------------------			
Study 1 7-3-86						
CCA[3] & PA[4]						
@ 25 lbs/T	11:45 am	22	1.0	1.8	0.8	3.6
PA at 20 lbs/T	2:30 pm	24	1.5	2.2	1.6	5.3
CCA	4:00 pm	16	1.7	2.9	1.3	5.9
Untreated	5:20 pm	20	1.7	2.3	2.8	6.8
Untreated	9:20 pm	22	0.5	1.0	3.5	5.0
PA at 20 lbs/T	9:20 pm	22	0.5	1.0	2.1	3.6
Study 2 8-4-86						
CCA	2:00 pm	13	1.7	3.3	1.7	6.7
CCA and PA						
@ 25 lbs/T	9:15 pm	18	0.5	0.9	2.8	4.2
CCA	9:15 pm	17	0.5	0.9	4.6	6.0
Hay after rain	3:20 pm	13	--	--	0.2	40.0
(8-11-86)						
Study 3 8-21-86						
PA at 27 lbs/T	11:59 am	32	0.7	1.6	7.1	9.4
PA at 15 lbs/T	3:20 pm	26	1.0	2.6	4.1	7.7
Untreated	5:20 pm	25	1.5	2.9	3.1	7.5
PA at 30 lbs/T	8:00 am	29	0.5	0.6	6.6	7.7

1 Estimated value based on data collected from other research.
2 Actual measured losses.
3 Supplied by American Farm Products, Ypsilanti, MI and applied at a rate of 5 lbs of product/ton of dry matter. This product contains approximately 50% sodium carbonate and 50% potassium carbonate.
4 Supplied by American Farm Products, Ypsilanti, MI. The active ingredients are a mixture of 80% propionic and 20% acetic acids.

NITROGEN TRANSFER FROM FORAGE LEGUMES TO GRASS IN A SYSTEMATIC PLANTING DESIGN

L.S. BROPHY, G.H. HEICHEL, AND M.P. RUSSELLE

INTRODUCTION

Nitrogen (N) transfer is the movement of N from one plant to another. Because symbiotic N_2 fixation represents an input of N to a legume-nonlegume plant association, the transfer of legume-fixed N to a nonlegume growing nearby is of particular interest. Unlike the mineralization of N from soil-incorporated green manure, N transfer is thought to occur during the growth of the legume. Thus, N transfer from legumes to nonlegumes may partly substitute for N fertilizers in intercrops, pastures, and relay crops composed of simultaneously growing legume and nonlegume species. Estimates of transfer range from 26 to 154 kg N ha^{-1}, depending upon species composition of the sward or intercrop, productivity and duration of crop growth.

These studies used difference methods or yield determinations to demonstrate the amount and benefit of N transfer, but do not distinguish N transfer from other factors that increase N yield or dry matter yield, such as competitive effects on root distribution and soil N uptake capability, or changes in soil biology and chemistry.

The ^{15}N isotope dilution technique, in which the soil is labeled with ^{15}N, is potentially a more accurate method for study of N transfer. Dilution of the ^{15}N concentration in grasses grown in mixture with legumes suggests N transfer from the legumes, since legume-fixed N_2 will be of natural ^{15}N abundance. Because the different rows provided evidence that N transfer occurred over a distance of at least 20 cm, and that isotope dilution technique detects only transfer of unlabeled (natural ^{15}N abundance) atmospheric N, it does not determine transfer of soil-derived N from legume to nonlegume.

The objectives of this study were to: (i) determine the amount of N transferred from alfalfa and birdsfoot trefoil to associated reed canarygrass; (ii) define conditions for N transfer, including interplant distance and species ratio; (iii) compare birdsfoot trefoil and alfalfa for N transfer; and (iv) determine the effect of grass proximity on legume N_2 fixation. Because of its advantages in separating competitive effects from N transfer, ^{15}N isotope dilution methodology was used in this study.

MATERIALS AND METHODS

The study was conducted on 1 m^2 field plots on a silt loam soil at the University of Minnesota Rosemount Experiment Station. An aqueous solution of 69.8 atom percent ^{15}N-labeled $(NH_4)_2SO_4$ was sprayed evenly on the plot surfaces on 26 April 1983 at a rate of 2.84 g m^{-2}, equivalent to 0.44 g ^{15}N m^{-2}. Surface soil was mixed uniformly within each plot to a depth of 15 cm to reduce volatilization of NH_3. Seedlings of 'Norcen' birdsfoot trefoil, 'Saranac' alfalfa, and 'Rise' reed canarygrass were grown for 6 weeks in sand benches under natural light in the greenhouse. Roots and tops

of all plants were trimmed to 7 cm before hand-transplanting on 12 May 1983. The experimental design was a randomized complete block with five replicates of each species mixture. The equidistant (7.5 cm between plants within and between rows) planting pattern within each plot was systematic, consisting of monospecific rows (6.5 cm apart) of grass and legume (either alfalfa or trefoil) interplanted to create a gradient from grass (right side, Figure 1) to legume (left side, Figure 1). The total population of legume plus grass was 188 plants m^{-2}. Maximum species mixing occurred in the center rows of the plot.

Herbage from each row in each plot was separately sampled and analyzed. Border rows (rows 1 and 15) and the outermost plants of the central 13 rows were not sampled. Herbage was removed to 7 cm height on 8 July, 1 August, and 1 September, 1983, at approximately 10% bloom of the legumes. Samples were dried at 60°C, weighed, and finely ground, and analyzed for N and ^{15}N. Nitrogen transfer and N_2 fixation were determined using the isotope dilution method. The control for these calculations was the grass row at 91 cm (second farthest from legumes, right side, Figure 1).

RESULTS AND DISCUSSION

The evidence from our experiment is the first to verify that transfer of fixed N_2 from alfalfa and birdsfoot trefoil to reed canarygrass occurs, and that the proportion of grass N obtained from transfer is high (68% from alfalfa; 79% from birdsfoot trefoil). Second, seasonal variations in relative isotope concentrations show that percent of grass N derived from transfer tended to increase as the season progressed. Variation of ^{15}N concentration among plants in maximum transfer occurred where the legume/grass ratio was greater than 1:1. Finally, legumes growing in mixtures with grasses derived a greater proportion of their N from symbiotic N_2 fixation than from soil N. At third harvest, N derived from symbiosis was higher (95% in alfalfa, 92% in trefoil) for legumes grown in mixture with grass than in monoculture (86% in alfalfa, 80% in trefoil).

This evidence has several practical implications. Because the legume/grass ratio allowing maximum N transfer was at least 1:1, N-deficient pastures may benefit from similar species ratios. N transfer was observed over a distance of at least 20 cm, corroborating other reports that show N transfer benefits for nonlegumes intercropped with grain legumes. However, further study of N transfer distances is needed; systematic designs like the one used in this investigation could increase the space and time efficiency of such experiments.
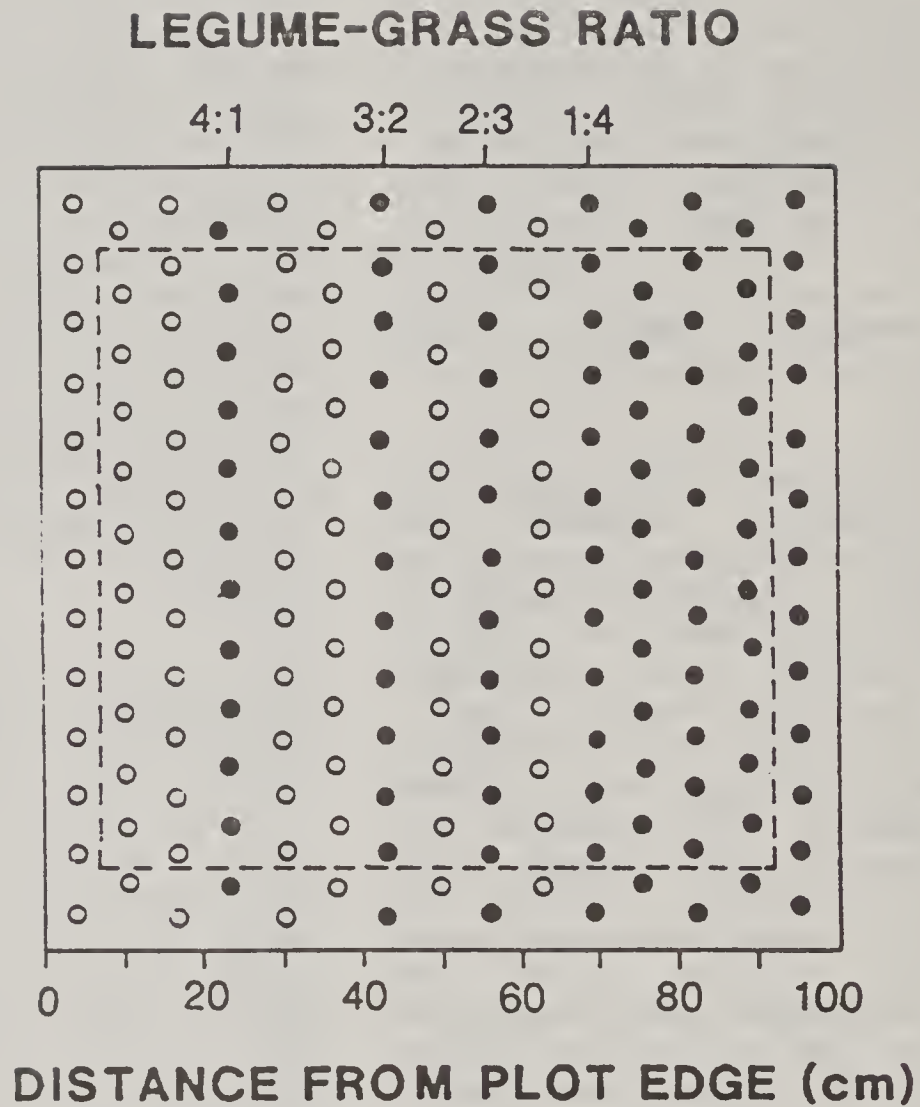
Figure 1. Systematic planting design for nitrogen transfer experiments. Rows of legumes are designated by open circles and rows of grass by closed circles. Spacings were 6.5 cm between adjacent rows and 7.5 cm between adjacent plants within and between rows. Sampling area is delineated by the broken line. The plot is legume dominant from distance 0 to about 50 cm, and grass-dominant from about distance 50 to 100 cm. The distances were measured from the left (east) edge of the plot.

NUTRIENT STRESS IN BIRDSFOOT TREFOIL
M.P. RUSSELLE AND R.L. MCGRAW

INTRODUCTION

Birdsfoot trefoil is widely grown as a forage legume in the North Central, Northeastern, and Pacific Coast regions of the United States, adjacent areas of Canada, and in many other temperate countries. Because it is better adapted than alfalfa to poorly drained, low fertility, and low pH soils, it is often grown under marginal management.

The responses of many crop species to nutrient stresses have been characterized, including several temperate and tropical forage legumes, but birdsfoot trefoil has received only scant attention. Knowledge of visual symptoms of nutrient stress can help producers and consultants diagnose the causes of declines in productivity and persistence. It is also important to know how nutrient stresses affect chemical composition, shoot and root growth and morphology, seed production, and nodule efficiency, because such effects may be present even when obvious visual symptoms are absent. Our objective in this experiment was to document several aspects of single nutrient stresses in one cultivar of birdsfoot trefoil. Norcen birdsfoot trefoil was used in this study because it is a high-yielding synthetic cultivar widely adapted to the northern United States and Ontario, Canada.

MATERIALS AND METHODS

Seeds were sown in acid-washed sand moistened with deionized water and were allowed to grow until the first true leaf stage. Seedlings were inoculated with nutrient-free, gnotobiotic solutions containing birdsfoot trefoil Rhizobia.

Four seedlings were transplanted to the glasshouse on 5 November in 7.5-L capacity PVC pots filled with nutrient solution for each of the treatments, which included deficiencies of P, K, Ca, Mg, S, B, Fe, Mn, Zn, Cu, and Mo. Concentrations of nutrients in the nonstressed treatment were similar to those used by Asher and Loneragan. Concentrations of specific elements required to induce visible symptoms were established in a short-term preliminary experiment. Nitrate-N was provided to reduce the impact on plant growth of expected nutrient stress effects on nodule formation and function. Pots were emptied, washed with deionized water, and filled with fresh nutrient solution weekly. Aeration was continuous and vigorous. Solution pH was maintained at 6.5 ± 0.6 through twice weekly measurement and was adjusted with NaOH and HCl. Sunlight was supplemented with fluorescent bulbs (125 μmol m^{-2} s^{-1}) for 14 h each day. Ambient air temperatures varied from 20 to 26°C. Plants were grown through two vegetative cycles and one reproductive growth cycle.

RESULTS AND DISCUSSION

Dry mass of shoots, shoot branching, and leaf area were typically lower in stressed plants than in plants grown in complete nutrient solution and specific leaf mass was generally larger in stressed plants. Nodule function and appearance were more severely affected by macronutrient than by micronutrient (except B) deficiencies. A deficiency or excess of a given nutrient often increased concentrations of other

nutrients, but concentrations of some elements also decreased in response to a stress in another. In a few instances, concentrations of a nutrient differed from the complete treatment in an opposite manner in shoots than in roots. Seed yield and yield components were less frequently affected by nutrient stress than were herbage yields. Visual symptoms were generally similar to those of alfalfa and the clovers.

In this experiment, we documented for the first time many of the responses of birdsfoot trefoil to single nutrient stresses. Clearly, each nutrient affects growth and development in different ways. This species appears to be less sensitive to Mn toxicity than other species, but may be somewhat more sensitive to high concentrations of Zn. It also appears that growth was reduced at substantially higher concentrations of P in solution than with tropical legumes, although

different experimental methodology may account for this discrepancy. Others have demonstrated that birdsfoot trefoil requires higher concentrations of P in soil solution than red clover or flat-pea (Lathyrus sylvestris).

We wanted to discover the general nature of the response of birdsfoot trefoil to insufficient or excessive amounts of several nutrient elements, and we were limited in the number of plants per treatment. Because of the variability among plants within a birdsfoot trefoil cultivar, more plants should be utilized in future experiments to estimate genotypic responses to specific nutrient stresses better. Cloned selections may be more useful for detailed physioloical experiments because plant-to-plant variability will be decreased. Details of nutrient concentrations and color photographs of visual symptoms are available from the senior author.

ESTIMATING N AND "ROTATION" EFFECTS IN LEGUME-CORN ROTATIONS

M.P. RUSSELLE, O.B. HESTERMAN, C.C. SHEAFFER, AND G.H. HEICHEL

INTRODUCTION

For centuries, farmers have known that yields of nonlegume crops are usually increased when they are grown simultaneously or in rotation with legumes. Yield increases of a nonlegume crop, such as corn, are usually attributed solely to the nitrogen (N) contribution by the legume, but positive effects not directly associated with N ("rotation effects") have been observed. These rotation effects

include improved soil physical properties, elimination of phytotoxic substances, and reduced disease incidence. Estimation of these rotation effects has been problematic and generally restricted to cases in which rotation systems continue to produce higher yields than monocrop systems after N limitations to yield have been removed by fertilization. The assumption in this approach is that

rotation effects are constant and not affected by changes in yield potential due to N availability. This assumption has not been tested to our knowledge.

Methods are needed to discern between N and rotation effects at any rate of applied N, so that reliable improvements in crop management systems can be made. Presumably, increased N contribution can be realized by use of appropriate legume harvest management or specially-developed cultivars, like 'Nitro' alfalfa. Without an understanding of the magnitude of rotation effects, little progress in their improvement can be expected. This paper reports one method which may be used to estimate N and rotation effects in crop rotations.

METHOD

This method is based on the relationship between grain yield and total N accumulation in the shoots of the nonlegume crop. Yield-N relationships are developed for continuous corn for the same year in which estimates of N and rotation effects are to be calculated to minimize confounding effects of environment. The total effect of a rotation is calculated as the difference in grain yield between corn after legume and corn after corn, at any specified rate of applied N. For a given legume treatment, a predicted value of corn grain yield is calculated from the total N accumulated in that treatment and the appropriate yield-N response function. The difference between the predicted yield and the actual yield of corn after corn is attributed to the improved N supply in the legume-corn rotation, and serves as an estimate of the N effect. The difference

between the predicted yield and the actual grain yield in the legume-corn rotation is interpreted as the rotation effect.

For example, assume that yields of continuous corn and corn after alfalfa at a particular rate of applied fertilizer N at the irrigated site were 103 and 150 bushels per acre, respectively, and that N accumulation in these treatments was 75 and 100 pounds per acre, respectively (Figure 1). The predicted yield of corn grown in rotation with alfalfa would be 132 bushels per acre, based on N accumulation alone. The total effect of rotation would be 47 bushels per acre (150 - 103), the N effect would be 29 bushels per acre (132 - 103), and the rotation effect would be 18 bushels per acre (150 - 132). The rotation effect represents 38% of the total effect in this example.

DISCUSSION

Two assumptions implicit in the proposed approach are, first, that the effects of N and non-N factors are additive and do not interact, and second, that the relationship between N accumulation in corn shoots and roots is either constant across rotation systems or does not significantly influence our estimates. Included in the N effects are all impacts of the cropping sequence on soil and crop residue N mineralization, indirect effects on N absorption by the nonlegume roots (for example, improved soil aeration), altered fertilizer N availability, etc. Our method cannot provide a measure of these individual processes, but we assert that the net effect of these direct and indirect effects on N availability is most completely described by the N accumulated in

the nonlegume. It should also be clear that the proposed method cannot differentiate among the various processes affecting the rotation component.

The proposed approach is the first which allows discrimination between N and non-N effects in crop rotations, without the use of N isotopes. It should be tested under a wide variety of conditions to determine its general utility, but should be applicable to any tillage system and to most crop rotation combinations.
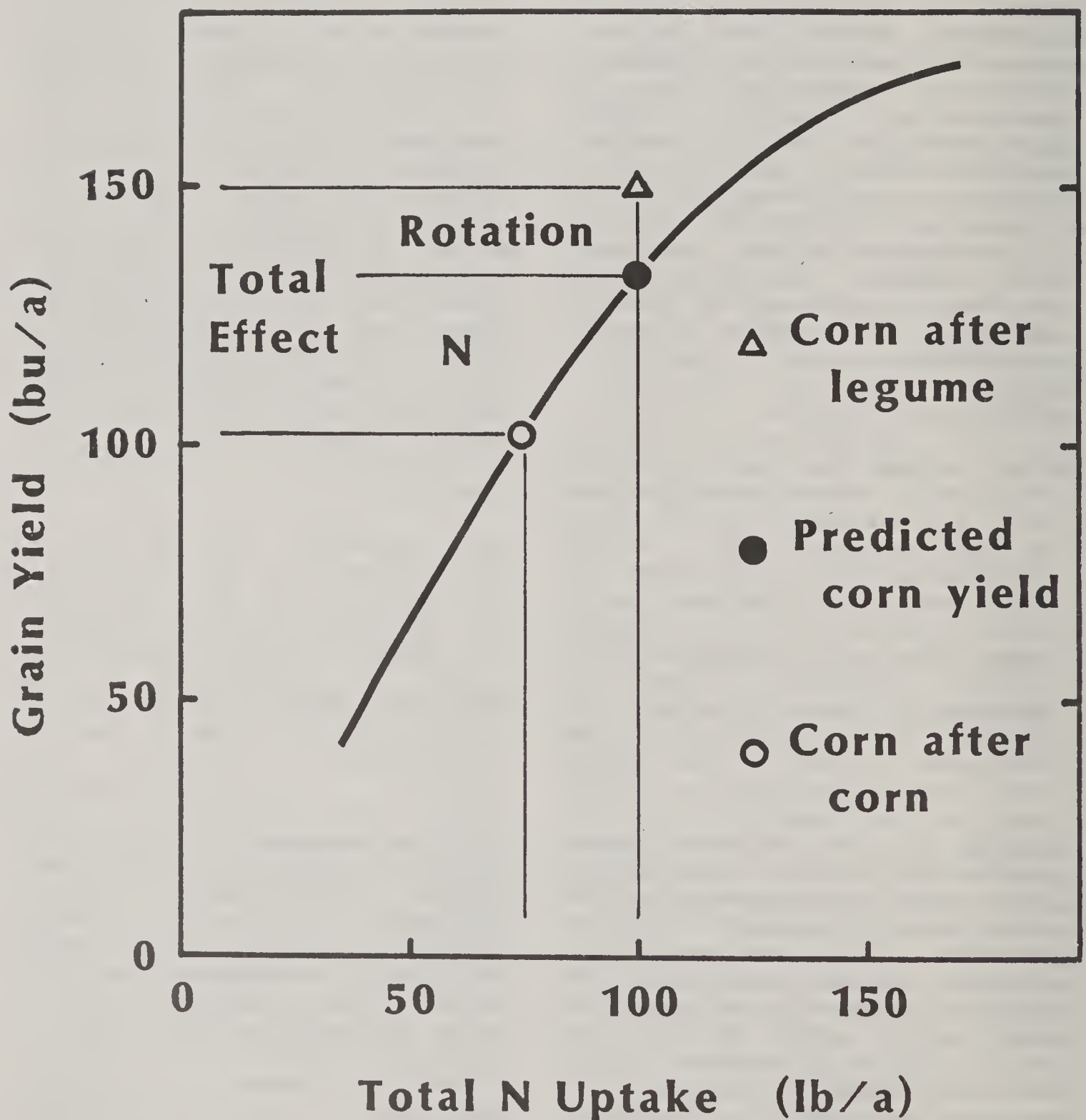

Figure 1. Relationship among total, N, and rotation effects in a hypothetical corn-alfalfa rotation.

IMPROVING THE QUALITY OF FORAGE HARVESTED IN LARGE PACKAGES

R.G. KOEGEL AND R.J. STRAUB

The labor-saving characteristics of large bales have made them attractive. However, forage quality has frequently suffered due to both increased leaf shatter during baling and more extensive spontaneous heating (with resultant nutrient loss) relative to smaller conventional bales. Outdoor storage further exacerbates losses.

Mechanical losses during baling can be minimized by baling at moisture contents too high to permit conventional storage. This technique thus necessitates the use of chemical preservatives, finish drying, or ensiling under anaerobic conditions. All three techniques, when properly carried out, can limit losses due to spontaneous heating to acceptable levels. In all cases, information on proper techniques as well as on labor, energy, and material expenditures are needed.

I. RADIAL FLOW DRYING OF LARGE, ROUND BALES

Considerable research has been conducted on axial flow drying both in the U.S. and in Europe. Results show that variations in density and in moisture content from layer to layer cause nonuniform air flow and drying. Assuring that all locations in the bale are adequately dried necessitates over drying much of the bale. This wastes energy and time. Causing air to flow radially through the bale from a hollow center, forces it to flow through all layers regardless of differences in their condition. In addition, radial flow results in a shorter flow path than axial flow, and consequently requires less power to force air through the bale at a given flow rate.

Bales were formed with a hollow center concentric with the bale axis. This was done initially by forming the bale around a wire mesh tube which was placed in the bale chamber and subsequently by using an 8" diameter plastic tube which was then withdrawn from the finished bale before drying. During drying, the bale axis was vertical. A flexible air duct was attached from an axial flow fan to the hollow center of the bale. Air was prevented from flowing out the bale ends by a weighted plywood disk on top and by the floor on the bottom. Alfalfa was baled at moisture contents from 20% to 44%.

For a given fan pressure, average radial air flow through bales was 55% higher than axial flow. Bales could generally be dried to a storage moisture content in from 24 to 48 hours without addition of heat. Adding heat reduced drying time, but added significantly to energy costs. Air flow rate was closely proportional to fan pressure. However, increasing flow rate did not necessarily decrease drying time significantly. This indicates that most flow rates used during the study were excessive. Energy costs for fan operation ranged from $6.50 to $15.00 per ton of dry matter. Uniformity of bale density from end to end varied greatly. It was found that uniformity could be significantly improved by carefully controlling the width of windrow.

In alfalfa with large diameter stems, especially at higher moisture levels, there was a tendency for stems to flatten. This increased flow resistance especially in the radial direction due to a "shingle" effect.

Future research will attempt to define minimum air flow rates at which near maximum drying rates can be achieved for given air conditions. This would minimize fan energy costs.

II. SILAGE FROM LARGE ROUND BALES

Bales at approximately 60% moisture content were enclosed in 6 mil polyethylene film in three different configurations: (1) single bales in bale bags; (2) a single row of bales end to end wrapped in sheeting with the edges rolled together and covered with soil; and (3) a stack of bales with six in triangular cross-section, wrapped with sheeting which was protected by a reinforced polyethylene tarp. The stack configuration had advantages in quantity of plastic required, labor required for wrapping, and the surface area available for gas exchange, all on a per bale basis. Temperatures of each bale were monitored.

The stack covering was damaged by rodents after approximately two months. This was repaired and further damage was controlled by the use of electric fencing and repellent. However, based on the temperature history of the stack, extensive damage had already occurred. The covering on the other two configurations remained intact.

Based on the mold found in all bales, preservation was judged to be unsatisfactory in all configurations.

Further research is needed on the permeability of various types and thicknesses of plastic films. Based on available data, gas exchange can be relatively high with the types of plastic film frequently used. Another requirement for this type of storage is absolute control of rodents.

REDUCTION IN THE ENERGY REQUIREMENT OF FORAGE CHOPPING

R.G. KOEGEL, F.J. FRONCZAK, K.J. SHINNERS AND R.J. STRAUB

Reduction in the specific energy required for forage chopping, in addition to reducing costs, could lead to more timely completion of harvesting for a given size of power unit. Previous work has shown that significant reduction of the cutterhead energy requirement is possible.

Initial work carried out in an instron testing machine showed that a cutting system based on a knife and anvil rather than the conventional knife and shear bar had energy requirements which compared favorably with the latter. It was further determined that bending the forage prior to cutting to put

tension on the knife side of the cut served to reduce the energy requirement of cutting. This benefit, however, was largely nullified when the energy required for bending was taken into account.

Based on the relatively low energy requirement found for knife and anvil cutting and the potential for reducing kinetic energy and windage losses relative to existing equipment, a rotary cutterhead was constructed to allow further study of the process. This cutterhead consisted of two counterrotating cylinders with alternate knives and anvils extending radially from their surfaces. The rotation of the two cylinders was timed so that the knives of one cylinder conincided with the anvils of the other. When the rotors were preloaded against each other and rotated, forage passing between them was cut between knives and anvils. The theoretical length of cut was 1/2 inch.

Some conclusions reached on the cutterhead were:

1. A cutterhead rotational speed of at least 600 rpm (peripheral speed of 14 miles/hr) was desirable to help clear rotors by centrifugal force.

2. High rotor to rotor preloads and a frame of high stiffness was required for clean cutting and satisfactory average length of cut relative to theoretical length. Average length of cut varied frm 0.4 to 1.3 inches.

3. Net cutting energy requirements ranged from 0.15 to 1.84 hjp hr/ton d.m. Comparable energy requirements of conventional equipment from the literature range from 0.37 to 1.2.

4. The maximum throughout for the 8" diameter by 8" long rotors was approximately two tons of dry matter per hour.

QUICK-DRYING FORAGE MATS

R.G. KOEGEL, K.J. SHINNERS AND R.J. STRAUB

Mats made from alfalfa shredded at the time of mowing and placed on the stubble have been shown in earlier research to dry to a moisture content suitable for baling in from 2 1/2 to 6 hours under favorable conditions.

A research prototype was constructed which concurrently carried out four functions: (1) mowing, (2) macerating (shredding), (3) forming into mats, and (4) placing mats onto the stubble. The cutterbar width of the machine was 120 cm making it smaller than a farm-scale machine. The macerator and press widths were 71 and 61 cm, respectively. This machine was operated, evaluated, and modified as necessary during the summer of 1986. It demonstrated the feasibility of performing the four functions concurrently in one machine.

Specific Energy Requirements, Capacity and Resulting Material Physical Parameters for Cylinder and Roll Macerator. (Cylinder and roll surfaces knurled.)

Parameter	Gross Specific Energy (kJ/kg d.m.)	Net Specific Energy (kJ/kg d.m.)	Capacity (Mg d.m. /hr)	Surface Area Index	Long Fiber Index
Cylinder : Roll Speed Ratio	(Eight rolls; Cylinder Speed 1100 rpm; Material Moisture 72% w.b.)				
* 1.3:1	24.4a	17.9a	2.19a	117.3a	0.408a
1.5:1	36.3b	24.9b	1.91b	130.7b	0.282b
2.0:1	37.9b	28.4c	1.46c	126.4b	0.333b
2.5:1	41.0c	28.1c	1.13d	132.4b	0.312b
Number of Rolls	(Speed Ratio 1.5:1; Cylinder Speed 1100 rpm; Material Moisture 79% w.b.)				
Eight	38.8a	25.9a	1.82a	139.7a	0.364a
Seven	32.0b	20.5b	2.00b	134.4b	0.364a
Six	30.1c	17.9c	2.19c	133.1b	0.377a
* Five	27.1d	15.9d	2.37d	131.9b	0.391a
Moisture Content (wet basis)	(Speed Ratio 1.5:1; Cylinder Speed 1100 rpm; Eight rolls.)				
71.9%	38.6a	32.2a	2.18a	122.5a	0.416a
57.0%	44.6b	37.1b	1.85b	112.5b	0.392a
50.0%	66.3c	54.3c	1.17c	106.9c	0.418a
38.9%	43.2b	33.5a	1.44d	104.1c	0.451a
Cylinder Speed	(Speed Ratio 1.5:1; Eight Rolls; Material Moisture 72% w.b.)				
500 rpm	39.5a	30.9a	0.86a		
650 rpm	31.7b	23.9b	1.19b		
800 rpm	33.1b	26.2b	1.68c		
1100 rpm	31.3b	25.6b	2.43d		

Note -- Average values with different markers are significantly different at the 95% level.

-- * These machine configurations produced material with acceptable physical parameters within the desired energy and capacity requirements.

Areas for future work include improvement of the uniformity of the mat thickness and reduction in press complexity, weight, and friction horsepower.

More extensive studies were carried out on the maceration of alfalfa to obtain relationships between the following independent parameters: (1) number of nip points, (2) surface speed ratios at nips, (3) rotational speeds, and (4) alfalfa moisture content and the dependent parameters: (1) resultant specific surface area, (2) weight percentage of "long fibers", (3) maximum machine throughput, and (4) specific energy requirements. Some results are given in the appended table. For more detail see Shinners (1986).

EPIPHYTIC LACTIC ACID BACTERIA ON ALFALFA PRIOR TO ENSILING

R.E. MUCK AND P.L. O'CONNOR

INTRODUCTION

Lactic acid bacteria are the major group of microorganisms responsible for the fermentation in the silo which preserves the ensiled crop. In order to insure that there are adequate numbers of lactic acid bacteria for a rapid fermentation, inoculants (most of which are specific strains of lactic acid bacteria) have become very common additives to alfalfa silage in the United States. Trials using inoculants in making alfalfa silages, however, have shown mixed success. One potential reason for an inoculant not providing a more rapid fermentation is that the natural or epiphytic population of lactic aid bacteria may have been higher than the number added by the inoculant. The objective of this study was to determine if there were factors which a farmer could easily assess which would allow estimation of the number of epiphytic lactic acid bacteria on alfalfa prior to ensiling.

MATERIALS AND METHODS

In both 1985 and 1986, we sampled alfalfa from alternate wagon loads as they were emptied into silos at the U.S. Dairy Forage Research Center at Prairie du Sac. These samples were analyzed for lactic acid bacteria and moisture level. Weather, equipment utilized in harvesting, length of wilting and dry matter yields for each cutting from each field were recorded and correlated with bacterial numbers. During second and third harvest 1985 and first harvest 1986, random samples of alfalfa before and after mowing were taken for microbial analysis. Samples were also taken from the top and bottom of swaths immediately prior to chopping during the second and third harvests of 1985. Sampling of the undersides of swaths was expanded in 1986. A minimum of two swath samples per field harvested per day were taken. Soil moisture, swath depth and swath

width on the day of harvest were also measured for each field in 1986.

RESULTS AND DISCUSSION

On fresh standing alfalfa, there were insignificant numbers of lactic acid bacteria (<10 CFU/g alfalfa). Mowing caused a significant increase in the bacterial level in approximately half the samples. There were no significant differences in the amount of inoculation between types of mowing machines (sicklebar or rotary). Consequently, any large number of epiphytic microorganisms on alfalfa entering the silo appeared to be the result of microbial growth during wilting or from inoculation during harvesting.

Analysis of both the swath and load data indicated that temperature during wilting was the most significant factor related to bacterial counts. In the swath very little microbial growth was evident when wilting temperatures were below 15°C. Above that, numbers tended to increase expontentially with temperature. Similarly in the load samples, high counts were always associated with high wilting temperatures. However, as seen in Figure 1, high wilting temperatures did not guarantee high populations of lactic acid bacteria.

The second most important factor was swath depth or indirectly dry matter yield. For depths above 5 cm or yields greater than 2 t DM/ha,
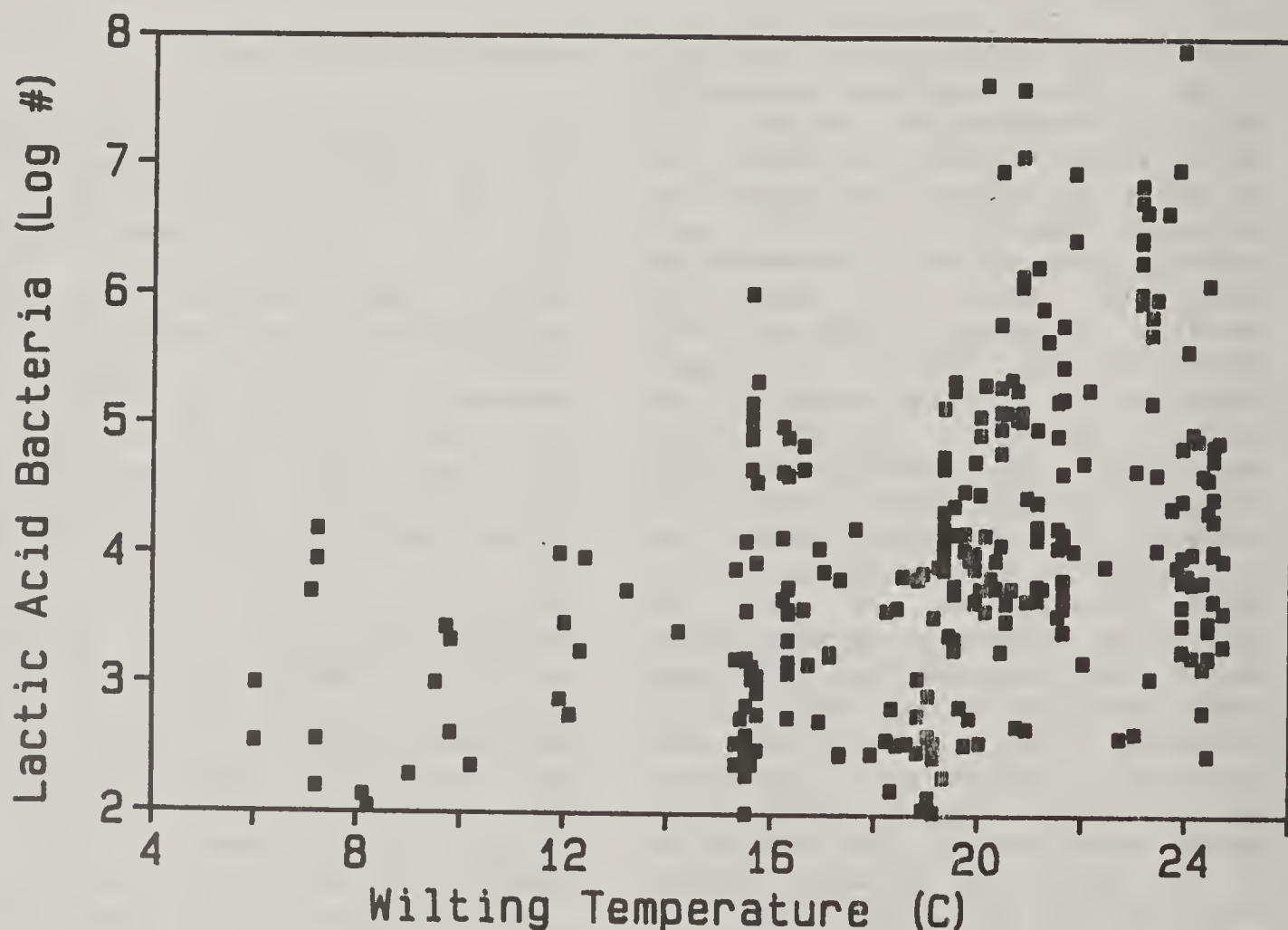

Figure 1. Lactic acid bacterial counts on harvested alfalfa as correlated with wilting temperature.

numbers of lactic acid bacteria on load samples increased with increasing depth or yield. Below those levels, epiphytic counts from load samples were generally less than 10^4 CFU/g alfalfa regardless of temperature. Analysis of the swath samples, when swath depths were below 5 cm, showed that virtually all counts were below 100 CFU/g alfalfa, and most were close to or below the detectable level of 10 CFU/g.

These results pointed to the forage harvester as the third major determinant of lactic acid bacterial populations on alfalfa entering the silo. When bacterial levels were low in the swath (<10 CFU/g) due to low temperature and/or low yield, the harvester boosted the population to between 100 and 10,000 CFU/g. The amount of inoculation increased exponentially in relation to the air temperature at the time of harvesting.

In addition to these three factors, wilting time and soil moisture during wilting were positively correlated with LAB counts; however, their effects were not as pronounced.

Based on these results, low epiphytic LAB counts under northern Midwest conditions would typically be expected in third (August) and fourth (October) cuttings and in years in which first cutting alfalfa is harvested under cool conditions. In these situations, an inoculant is most likely to show some benefit.

AEROBIC DETERIORATION OF ALFALFA AND CORN SILAGES

P. O'KIELY, R.E. MUCK AND P.L. O'CONNOR

INTRODUCTION

To properly preserve a forage as silage, it must be stored in an air-free environment, and the activity of anaerobic microorganisms must be inhibited. As long as these conditions prevail the silage will remain stable. Once silage feeding commences, the silage being fed and some of the silage in the silo will be exposed to air. The first fundamental requirement for silage stability has then gone, and the silage is potentially unstable. If unstable, quantitative and qualitative losses will occur, producing CO_2, water and heat as by-products. The experiments reported here determined the relative aerobic stability of alfalfa and corn silages and studied the inherent characteristics in alfalfa silage that confer aerobic stability.

MATERIALS AND METHODS

Four sets of experiments were conducted. In the first experiment, an apparatus was constructed that permitted CO_2-free air to be passed through silages held in 100 ml centrifuge tubes and that provided means for measuring temperature, CO_2 production and dry matter loss. A wide variety of silages were placed in the apparatus

for 7 or more days of aerobic exposure. Silages tested in this manner included: a farm-scale corn silage, a series of third-cut alfalfa silages made in test tube silos at 4 different dry matter levels (25 to 55% DM) with and without the addition of glucose at ensiling, a series of fourth-cut alfalfa silages made similarly, and 10 farm-scale alfalfa silages.

In the second experiment, micro-organisms present on several aerobically unstable and deteriorated alfalfa and corn silages were collected and grown on a media that would select only those that could grow aerobically on lactic acid at pH 3.5. This destabilizing inoculum contained predominantly yeasts. The previously studied silages were inoculated with these organisms at approximately 10^6 CFU/g silage and then tested for aerobic stability as in the first experiment.

To indicate if the stable alfalfa silages contained substances to inhibit fungal growth, corn silage, a stable alfalfa silage and a 50:50 mixture of both were all inoculated with the destabilizing inoculum and tested for stability as done previously.

A final experiment was performed to determine if the factor causing alfalfa silages to be stable was present in the herbage. Six varieties of alfalfa and one each of birdsfoot trefoil and red clover were harvested and a portion of each was ensiled in test tube silos for 16 days. Destabilizing inoculum was added to petri plates containing malt agar acidified to pH 3.5 with lactic acid. These plates served as controls. On similar plates, either fresh or ensiled herbage, with or without autoclaving, was added to the inoculum and agar. After

several days incubation, each plate was counted for yeasts and molds.

RESULTS AND DISCUSSION

In the first experiment, all of the test-tube alfalfa silages were extremely stable throughout 7 days of aeration. Dry matter losses, CO_2 production, heat production and pH change were negligible. Over the same period, the corn silage had a 16% loss of dry matter, a corresponding production of CO_2 and heat, and a pH increase from 3.9 to 8.1. Seven of the 10 farm-scale alfalfa silages were as stable as the test-tube silages; however, 3 of the 10 were unstable, averaging 6% dry matter loss in 7 days.

The addition of a destabilizing inoculum did not affect the stability of any of the stable alfalfa silages. Measurement of yeast numbers in these silages showed that yeast numbers often dropped rapidly within 24 h after inoculation. In contrast, the destabilizing inoculum caused more rapid and extensive aerobic deterioration in the silages which had been unstable in the first experiment. These results indicated that the number of destabilizing microorganisms in any of the stable alfalfa silages was not a factor affecting stability.

In the third experiment, the corn silage showed rapid heat production and dry matter losses while the alfalfa silage was extremely stable. The mixture had dry matter losses intermediate between the two. However, measurements of yeast numbers, pH and temperature with time showed a distinct lag before signs of instability in the mixture. These results implied the presence of a yeast inhibitor(s) in the alfalfa silage.

The fourth experiment indicated that the inhibitor(s) in alfalfa silage was not present in the herbage but was a product of fermentation. Non-autoclaved fresh herbage was associated with extensive mold development covering all plates. This was to such an extent that yeast growth was restricted. Autoclaving the herbage overcame this problem, but plates with autoclaved fresh herbage had similar yeast levels as the control plates, implying that the fresh herbage had no effect on yeast development. Yeasts on the plates containing fresh herbage were as likely to be positioned close to plant particles as not.

Plates containing non-autoclaved silages showed no mold growth, implying that the molds which were present on the fresh herbage did not survive ensiling. Due to yeasts indigenous to some of the silages, some of the plates from the non-autoclaved silages had elevated yeast counts compared with the controls whereas many had substantially reduced counts. With the autoclaved silages, all herbage plates showed reductions in yeast count over controls, averaging 50%. This was true of the trefoil and clover as well as of the alfalfa silages. On these plates, yeast colonies were not randomly distributed but distinctly avoided the silage particles.

Consequently, these experiments indicate that many alfalfa silages are very aerobically stable and that this stability is due to a compound or compounds produced during fermentation which is (are) inhibitory to fungal growth.

ALFALFA QUALITY AS INFLUENCED BY MECHANICAL AND CHEMICAL CONDITIONING

C.A. ROTZ, R.J. DAVIS AND S.M. ABRAMS

INTRODUCTION

Conditiong is a process which speeds the field drying of forage crops. Mechanical methods of conditioning by crushing, breaking and abrading the forage material are widely used by forage producers. The most popular mechanical treatment is the use of intermeshing rubber rolls which both crush and break the stem, facilitating the movement of water out of the plant. A relatively new method of conditioning alfalfa uses chemicals which affect the waxy surface of the plant, allowing moisture to leave the plant more easily. Spraying a 2.8% aqueous solution of potassium carbonate on the crop as it is mowed provides the most effective chemical conditioning treatment.

Research has shown that both types of conditioning can speed the field drying of alfalfa and thus reduce the field curing time. The ultimate goal, however, is to produce higher quality hay, and the relationship between field curing time and hay

quality has not been well documented. The objective of this work was to monitor alfalfa quality during field curing and storage to determine differences created by the method of conditioning.

MATERIALS AND METHODS

Alfalfa was mowed with either a mower or mower-conditioner which used a reciprocating cutterbar for mowing with a 2.8 m (9ft.) width of cut. The mower-conditioner included intermeshing rubber rolls for conditioning, and a maximum roll pressure was used for all tests on all cuttings. To provide chemical conditioning, a spray boom with a push bar was mounted on the right side of the tractor to spray the standing alfalfa just before it was mowed. The push bar opened the plant canopy to allow the spray to penetrate to the lower stems. The chemical applied was a 2.8% solution of potassium carbonate in water.

The experimental design included 16 replications of the same test over a wide range of crop and environmental conditions during a two-year period with three cuttings per year. Each test included four conditioning treatments: 1) none, 2) mechanical, 3) chemical and 4) mechanical and chemical combined. To eliminate influence from variations in crop and soil conditions, swaths of alfalfa were mowed with the four treatments in random order, side-by-side in the field. When the alfalfa of each treatment reached a target moisture content of 20% (wet basis), it was baled with a conventional retangular baler and 10 bales of each treatment were stacked in a barn for storage.

Hay quality was monitored for each treatment from the standing crop through storage. Three samples were obtained from each treatment immediately after mowing and at the end of each field curing day by randomly gathering and chopping 500 g of material per sample. Samples were obtained in and out of storage by coring three bales of each treatment. Freeze drying was used for sample preparation.

Quality was measured using Near Infrared Reflectance Spectroscopy (NIRS). Quality measurements were crude protein (CP), water insoluble nitrogen (IN), in vitro dry matter disappearance (IVDMD), acid detergent fiber (ADF), and neutral detegernt fiber (NDF). Approximately 10% of the samples were randomly selected for a standard laboratory analysis to provide data for calibration of the NIRS unit.

RESULTS AND DISCUSSION

Significant differences in hay quality were not found among the conditioning treatments throughout the field curing process; however, quality differences were found as hay was placed into and removed from storage. For hay placed into storage, only minor differences in quality were measured. CP, NDF, ADF and IVDMD were not affected by conditioning treatment. More insoluble nitrogen was found as the amount of conditioning increased.

A significant trend in quality was found between treatments in hay removed from storage. Overall, unconditioned hay was higher in fiber content when compared to conditioned hay. This difference can be attributed to microbial consumption of readily available energy sources in the cell solubles fraction (non-NDF fraction) of unconditioned hay, such as soluble carbohydrates, organic acids, starch

and pectin. Microbial activity occurred because unconditioned hay was baled at a higher moisture content which was conducive to microbial activity during storage. The lower content of cell solubles was also reflected in the lower IVDMD of unconditioned hay. The higher levels of insoluble crude protein in unconditioned hay, expressed as a percentage of total crude protein, suggests either conversion of forage nitrogen into less soluble microbial protein, or creation of bound nitrogen by the Maillard reaction due to heating of the hay during storage. The slightly higher concentration of crude protein in unconditioned hay compared to chemically conditioned hay was most likely due to the loss of cell solubles in unconditioned hay, which resulted in protein, as

well as fiber, making up a larger fraction of total forage dry matter.

Although conditioning treatments have a large effect on drying rate, they do not effect hay quality when exposure to precipitation and moisture content at baling are not influenced by treatment. Rain damage can greatly reduce hay quality. When the use of conditioning allows rain damage to the crop to be avoided, quality differences will occur. This, however, never occurred in this series of tests. Unconditioned hay will often be baled at a higher moisture content because of slow drying. Higher moisture levels in hay entering storage will cause greater loss of quality during storage.

CHEMICAL CONDITIONING OF ALFALFA WITH CONTROLLED DROPLET ATOMIZERS

C.A. ROTZ AND R.J. DAVIS

INTRODUCTION

Chemical conditioning is a relatively new concept for speeding the field drying process in alfalfa hay production. Chemical conditioning is performed by spraying the crop with an aqueous solution of 2.8% potassium carbonate as it is mowed. The chemical affects the waxy cutin of the plant to allow moisture to leave the plant more easily.

When conventional spray equipment is used to apply the treatment, large volumes of water are applied to

assure good coverage and rapid drying of the alfalfa. Mixing and handling of the chemical at high application rates will slow the mowing operation and increase labor and fuel requirements. If application rate were reduced without sacrificing treatment performance, chemical conditioning would be more practical and perhaps more economical for the farmer.

Controlled droplet atomizers (CDA's) are being used to apply chemicals on agricultural crops at a reduced

carrier application rate. The objective of this work was to determine if CDA's could be used in chemical conditioning of alfalfa to reduce the required amount of water carrier while maintaining plant coverage and drying rate.

MATERIALS AND METHODS

Field drying experiments were conducted over a 4-year period. Each experiment included 6 or more individual field tests on different dates and under a variety of conditions. Both the relative amount of coverage the alfalfa received with each treatment and the resulting drying rate were measured. Four types of CDA's were evaluated which included a rotary atomizer and two types of air-carrier CDA's. The centrifugal atomizer used a spinning disk to break up the solution and distribute it in a vertical plane. The unit was shielded to divert all spray material downward. The most extensively tested CDA used a centrifugal fan for breakup and distribution of the chemical. A similar CDA, referred to as the air-curtain sprayer, used a rotary atomizer with straight stream airflow created by stacked crossflow fans. Both air-carrier CDA's were installed on the front of the mower-conditioner to blow the spray material throughout the crop canopy just before the crop was cut. A fourth spray system was tested which used air nozzles to apply the spray inside the hood covering the blades of a rotary disk mower. The last three CDA's all used air as a carrier in substitute for water.

As a basis for comparison, treatments were included in all experiments where chemical conditioning was applied through conventional nozzles and where no chemical conditioning was applied. Treatments with the alternative spray devices included a range of application rates.

RESULTS AND DISCUSSION

Air-carrier type CDA's normally provided more uniform coverage of the alfalfa than obtained with either a conventional nozzle spray system or a rotary atomizer type CDA. Spray coverage on the backside of alfalfa sprayed with a conventional nozzle was 30 to 50% less than that on the front of the plant. With the air-carrier CDA, coverage was similar on both sides of the plant with less variation from the bottom to the top of the plant.

Alfalfa drying rates were similar following all spray systems with drying rate directly related to the application rate of the 2.8% aqueous solution of potassium carbonate (Figure 1). The air-nozzle spray system on the rotary disk mower provided similar, but slightly poorer coverage and drying performance compared to the air-carrier type CDA's. Alfalfa drying rate was not affected by an increase in concentration of potassium carbonate or the addition of a surfactant in the chemical solution.

Although alternative spray devices may provide more uniform coverage of alfalfa with the chemical conditioning treatment, the improved coverage does not increase drying. These devices do not provide a practical means for more effective chemical conditioning. With little or no reduction in the amount of water required, the high initial cost of CDA equipment cannot be justified.
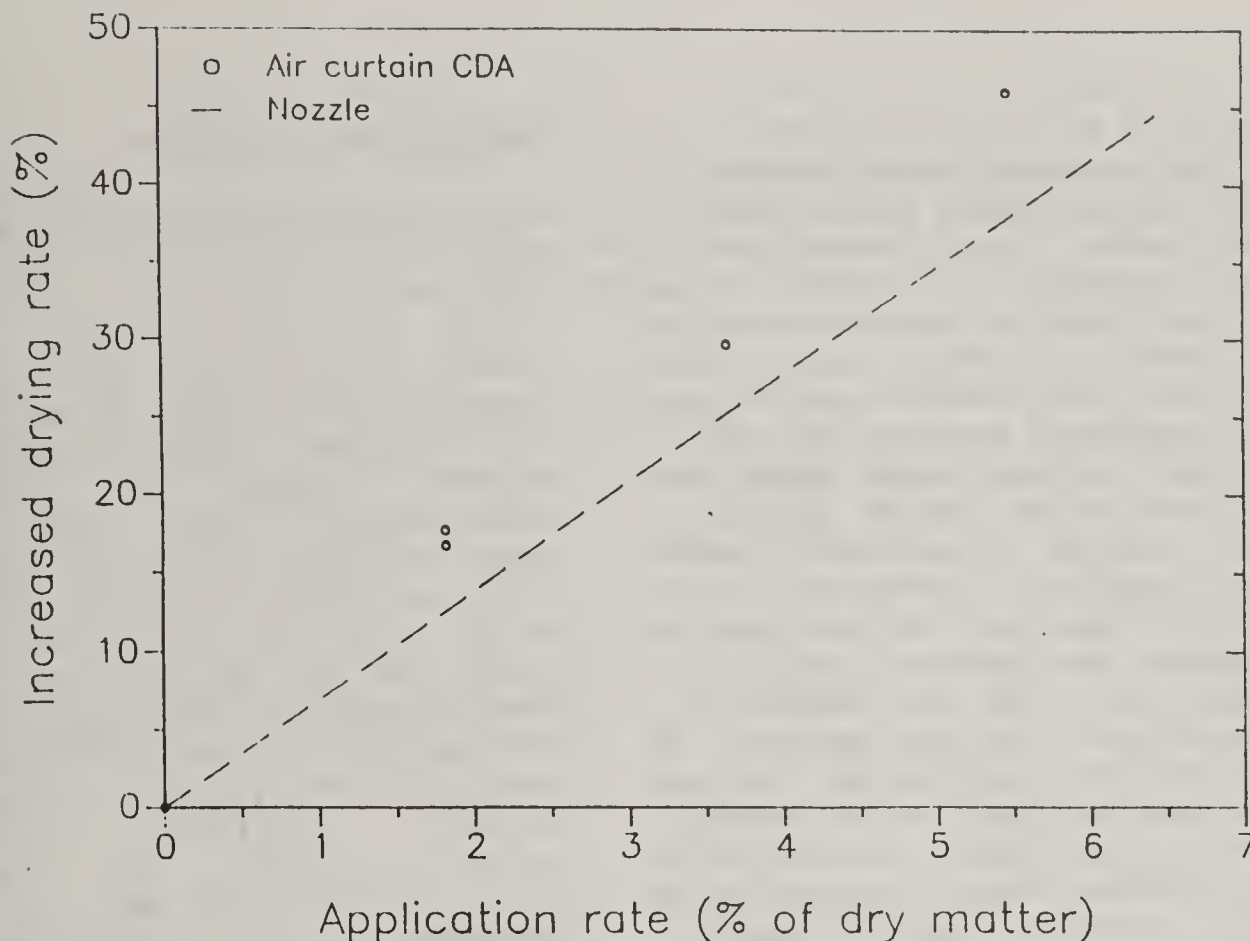
Figure 1. The increase in alfalfa drying rate obtained with chemical conditioning applied over a range of application rates using fan nozzle and air-curtain CDA spray systems.

EFFECT OF K AND CA FERTILIZATION OF ALFALFA ON RATE OF PH DECLINE AND MICROBIOLOGY OF RESULTING SILAGE

W.L. SHOCKEY

INTRODUCTION

The attainment of low forage pH is essential for preservation of crops as silage. Rapid rates of pH decline are especially important when ensiling high protein forages such as alfalfa because proteolytic enzyme activity is not inhibited until pH falls to 4.0 to 4.5. The K and Ca salts of organic acids could play a role as buffers in slowing the rate of pH decline. Fertilization practices can greatly affect the content of organic acids and inorganic ions in herbage used for silage. Objectives were to determine effect of K and Ca fertilization of alfalfa on rate of pH decline and microbiology of resulting silage.

MATERIALS AND METHODS

Four plots of alfalfa (9.8 x 45.7 m) were established in 1984 and fertilized with 840 (HK) or 0 (LK)

kg K/ha as KCl and 8,070 (HCa) or 0 (LCa) kg Ca/ha as $CaCO_3$ applied in 3 split applications before plowing, after plowing, and in March 1985 in a 2X2 factorial. In 1985, forage from the third cutting of each plot was packed into three 200 l steel drums lined with double plastic bags and evacuated. Samples for pH and microbial analyses were taken when drums were filled and on days 1, 4, 7, 21, and 60 drums were opened, core sampled, evacuated and resealed. Rate of pH decline was calculated by: pH = b (ln days) + a, where pH = pH of sample, b = slope or rate of pH decline, ln days = natural logarithm of the days after ensiling and a = constant. Number of anaerobic microorganisms were estimated with roll tubes after serial dilution under anaerobic conditions.

RESULTS AND DISCUSSION

Silage pH on days 1 and 60 and slope were 5.35, 4.54 and -.200; 5.41, 4.93 and -.134; 5.62, 5.21 and -.113; and 5.55, 5.08 and -.111 for HK-HCa, HK-LCa, LK-HCa and LK-LCa, respectively. Rate of pH decline was greatest for HK-HCa. The highest number of anaerobes (anaerobes/10^{-6} gm wet forage) found in silages from each treatment group were 580 on d 4 for HK-HCa, 37 on d 60 for HK-LCa, 97 on d 4 for LK-HCa and 101 on d 21 for LK-LCa. Results suggest that faster rate of pH decline for HK-HCa forage may have been the result of stimulated microbial activity. Microbial activity could be stimulated directly by the presence of higher concentrations of alkaline minerals or indirectly by differences in plant chemical composition, such as soluble sugars, caused by fertilizer treatment.

EFFECTS OF MICROBIAL INOCULANT ON FERMENTATION OF POOR QUALITY ALFALFA

W.L. SHOCKEY, B.A. DEHORITY AND H.R. CONRAD

INTRODUCTION

Previous experiments have found variable response to the addition of a microbial inoculant to forages. One reason for lack of effect is that most experiments were carried out using high quality forages ensiled under ideal conditions. In practice, a considerable amount of forage is harvested under less than ideal conditions. Objectives were to determine if the addition of microbial inoculant could enhance the fermentation of alfalfa that had been weathered for several days after cutting and before harvest.

MATERIALS AND METHODS

Second cutting alfalfa was cut at the midbloom stage and allowed to lay in the field for 10 d. Rain fell at least 4 of those days. Forage was chopped (2 to 4 cm) when the dry matter content reached 50%. Replicated, well-mixed loads of forage were halved and transferred to a feed mixing wagon and the microbial inoculant applied to one-half the forage. This application rate resulted in 7,000 to 10,000 viable anaerobes applied to each

gram of treated forage. After mixing (15 min), forage was packed into seven 200 l steel drums lined with double plastic bags (4 mil) and evacuated with an industrial vacuum cleaner. Forage samples were taken when drums were filled (d 0). On days 1, 4, 7, 21 and 60, drums were opened and duplicate samples taken for analysis of dry matter, nitrogen (total, nonprotein, ammonia), acid detergent fiber, pH, total nonstructural carbohydrate, lactic acid and acetic acid. Microbiological analyses included total (media pH = 6.8) and acid tolerant (media pH = 4.5) anaerobes and presumptive genus identification.

RESULTS AND DISCUSSION

Despite the deteriorated condition of the forage; i.e. low total nonstructural carbohydrate, high buffering capacity, and high ammonia; sufficient end products were produced to lower forage pH to 4.5 to 5.5. Production of ca. 8% fermentation acids from 4 to 5% nonstructural carbohydrate indicate that substrate sources other than those measured as total nonstructural carbohydrate, such as 5-carbon sugars and organic acids, contributed to overall end product formation. Treatment did not affect rate of acid production or production of nonprotein nitrogen and ammonia nitrogen, which are indicative of rates of proteolysis.

Total anaerobes were higher four d after ensiling in the inoculated vs control silage (45.0 vs 12.8 x 10^8 anaerobes/gm wet forage) indicating that growth of microorganisms was faster in the inoculated drums early in the fermentation. However, acid tolerant anaerobes were higher in the control vs the inoculated silage (11.2 vs 5.4 x 10^7 anaerobes/gm wet forage averaged over all sampling days) which suggests differences in the metabolic characteristics of the microbiological populations that developed. Even though differences in the development of the microbial populations were apparent, lack of treatment effect on silage chemistry suggests little difference in performance of animals consuming the silage.

EFFECT OF METHOD OF ENUMERATION AND STERILIZATION ON ANAEROBE NUMBERS OF FORAGES

W.L. SHOCKEY AND B.A. DEHORITY

INTRODUCTION

Quantification and characterization of anaerobic bacteria present on forages by the roll tube technique is a time consuming process. Most probable number procedures make it possible to estimate the number of bacteria and the characteristics of the major portion of bacteria present in a shorter amount of time. Further, studies of forage microbiology could be enhanced by the availability of sterile forage on which studies of fermentation reconstruction could be based. Objectives were to compare the enumeration of forage anaerobic

bacteria by most probable number or roll tube procedures and to test the efficacy of sterilization of alfalfa by exposure to ethylene oxide.

MATERIALS AND METHODS

Alfalfa, alfalfa/grass and corn silages; and alfalfa and orchardgrass herbage were used to compare number of total (media pH = 6.8) and acid tolerant (media pH = 4.5) anaerobes present on 13 forage samples using roll tubes or most probable number (MPN) procedures. Fresh cut alfalfa was exposed to ethylene oxide from 12 to 36 hours to determine time required to sterilize wet herbage. Sterilization was confirmed by lack of growth of bacteria after ensiling for at least 7 days.

RESULTS AND DISCUSSION

Concentrations per gram wet forage for total anaerobes were not different (overall means = 10.6 vs 16.0 x 10^7) but concentrations of acid tolerant anaerobes were higher (overall means = 15.3 vs .6 x 10^6) using MPN compared to roll tubes, respectively. Apparently, acid tolerant anaerobes proliferate more readily in a liquid medium. As determined by reduction in number of total anaerobes (87,400; 13,200; 560; and 185 microbes per gram wet herbage for 0, 12, 24, and 36 hours exposure to ethylene oxide, respectively), and by inability of microorganisms to increase in numbers after ensiling for 7 days (>18,000,000; 350; and 550 microbes per gram wet silage for 12, 24, and 36 hours exposure to ethylene oxide, respectively), 24 hours exposure to ethylene oxide was sufficient to sterilize forage. The technique allows preparation of sterile forage for studies of fermentation reconstruction and endogenous plant reactions.

A MODEL OF ALFALFA HAY STORAGE

D.R. BUCKMASTER, C.A. ROTZ AND D.R. MERTENS

INTRODUCTION

Alfalfa hay experiences dry matter loss and quality change during storage, particularly at elevated moisture contents. These losses reduce feed quality and decrease the amount of feed available to the dairy animal. Information on hay storage losses is not well documented. The objective of this work was to describe the changes which occur to baled alfalfa hay during storage by developing models which predict storage temperature, dry matter loss and changes in quality parameters as functions of the moisture content and density at the time of baling. The model considers losses during inside storage only; losses due to handling and/or weather exposure are not included.

MATERIALS AND METHODS

Models were developed and validated with experimental data. Three similar experiments were conducted; one each on first, second and third cutting alfalfa, 1985. Hay was baled at target treatments which included two levels of density and 6 levels of moisture content. Five bales of each treatment which appeared uniform in moisture and density were chosen to be monitored in storage.

Initial sampling was performed within two hours of baling. Core samples from each bale were oven-dried to determine moisture content or air-dried for quality analysis. Standard wet laboratory procedures were used to determine crude protein (CP), ash, acid detergent fiber (ADF) and acid detergent insoluble protein (ADIP) contents. Stack temperature of each treatment was monitored during storage. After 60 days, bales were removed from storage and again sampled to determine moisture content and quality.

Heat generated within the hay was determined by performing an energy balance on the stack. Heat generation rate was calculated from temperature data by assuming rates of heat and moisture transfer through the stack. Empirical relationships were developed which predicted heat generation rate as a function of time from baling, moisture content and bale density. These relationships, used in combination with a heat transfer model, can predict storage temperatures in a hay stack of any size or shape. A dry matter loss model was developed based upon the theoretical relationship of the chemical change due to microbial respiration and the energy balance on the hay stack. Models which predict changes in protein, ADF and ash were developed and validated by comparing predicted and actual quality changes for data collected from hay storage experiments.

RESULTS AND DISCUSSION

Models which resulted from the analysis are given in Table 1. Dry matter loss is a function of the heat generated in the stack and moisture content of the hay. Quality changes are functions of dry matter loss.

Dry matter loss was primarily soluble carbohydrate so the total amounts of ADF and ash remained constant during storage. Microorganisms may consume protein just as they consume carbohydrates; however, because carbohydrates are more soluble, CP is consumed at a slower rate. Experimental data indicated that CP was lost at approximately 40% of the rate at which other dry matter was lost (equation 4, Table 1).

A factor which affects protein availability in hay is protein bound to fiber when heating occurs. Linear regression analysis of experimental data indicated that bound protein (ADIP) increased in proportion to heating in degree days greater than 35°C (95°F). When bound protein exceeded 10% of crude protein, crude protein was discounted. Equation 6 in Table 1 contains a multiplicative discount factor for crude protein when the bound protein content becomes important.

The alfalfa hay storage model quantifies the changes which occur to hay during storage. The model has many useful applications for

predicting hay quality. The model is now part of a whole-farm simulation model of the dairy forage system where it will be used to evaluate alternative systems for hay harvest on the dairy farm.

Table 1. Alfalfa Storage Model.[a]

1. $\displaystyle DML = \frac{0.0355 T \times G_m + M_i - M_f}{5.84(1-M_i) - M_f}$

2. $\displaystyle ADF_{out} = \frac{ADF_{in}}{(1-DML)}$

3. $\displaystyle Ash_{out} = \frac{Ash_{in}}{(1-DML)}$

4. $\displaystyle CP_{out} = \frac{CP_{in}(1-0.4DML)}{(1-DML)}$

5. $\displaystyle ADIP_{out} = \frac{(ADIP_{in} + 0.00373DD)}{(1-DML)}$

6. If $\displaystyle \frac{ADIP_{out}}{CP_{out}} > 10\%$, discount CP_{out}:

$$CP_{out}, \text{ discounted} = CP_{out} \left(1.1545 - 1.545 \frac{ADIP_{out}}{CP_{out}}\right)$$

[a] Quality expressed as percent of dry matter. Subscripts indicate quality as placed into or removed from storage.
DML = dry matter loss (fraction of initial dry matter).
T = time of storage (d)
G_m = mean heat generation rate within the hay (W/kg).
M_i = initial moisture content (decimal wet basis).
M_f = final moisture content (decimal wet basis).
DD = degree days for which temperature exceeds 35°C.

PREDICTING INTAKE USING KINETIC CHARACTERISTICS OF NEUTRAL DETERGENT FIBER

D.R. MERTENS

INTRODUCTION

Last year a model for predicting intake was developed that was based on biological theories of intake regulation in animals. An equation was developed which describes the physiological energy demand theory that intakes (I) of diets with high energy contents (E) are regulated to meet energy requirements (R): $1 \times E = R$. The physical fill theory was described by an equation which indicates that intakes (I) of rations with high fiber content (F) are limited by daily fill capacity (C): $1 \times F = C$. Since E and F are inversely related these equations can be solved at their intersection to obtain the maximum dry matter intake that will meet the energy requirements of the cow: I_{max} occurs when $R/E = C/F$. The model was solved by using NDF to approximate the fill effect of the diet. It was recognized that both R and C were fluxes that could be expressed in terms of kinetic characteristics of the animal and feed. The objective of this project was to develop the kinetic equations for daily fill capacity and investigate effect of kinetic characteristics on expected feed intake by dairy cows.

MATERIALS AND METHODS

The kinetic model was derived by two different methods to verify the analytical solutions. The first method used simple substitution based on the kinetic definition of a flux to define C: Flux = (Pool) X (fractional rate constant). The second method involved the development of a simple kinetic model similar to that described by Mertens and Ely (J. Anim. Sci.

49:1085, 1979) and solving the model at steady-state to obtain intake as a function of the rumen pool of fiber and its turnover. After verification, the model was used to estimate intake for a typical alfalfa and orchardgrass forage using the kinetic parameter coefficients given in Table 1. The rumen pool of NDF was estimated to be 1.2 kg per 100 kg body weight based on preliminary analysis of animal data. To evaluate the effect of kinetic characteristics on intake each kinetic parameter was changed by 10% and the effect on fiber fill and intake was calculated.

RESULTS AND DISCUSSION

The two methods for deriving the kinetic model obtained the same result. Simple substitution indicated that daily intake capacity (C) of NDF was equal to the rumen pool (P) of NDF times its average rate of total disappearance (K_t): $C=(P)(K_t)$. Since the fill theory states that $I=C/NDF$ then $I=(P)(K_t)/NDF$. The analytical solution to the kinetic model at steady-state obtained: $I=P/(NDF)(TO_a)$; where TO_a is the average turnover time of all NDF in the rumen. The two derivations are equal because $K_t=1/TO_a$ in first-order systems.

The ranking of kinetic characteristics by their expected effect on intake is given in Table 2. The ranking of characteristics is not dependent on the estimate of rumen NDF pool size (1.2% BW); however, the value accurately predicted observed intakes and digestibilities.

The results suggest that NDF content and the potential digestibility of NDF have the greatest impact on intake of animals fed diets containing the maximum proportions of forage. Disappearance rates of fiber have important, but less dramatic effects on intake within a species of forage. The magnitude of the effect of rate of escape of fiber from the rumen indicates that factors affecting this characteristic deserve further investigation.

CONCLUSION

Adding kinetic characteristics to a theoretical model of intake regulation provides a method for developing research hypotheses and testing them quantitatively. Model simulations suggest that factors affecting potential digestibility of NDF and escape of NDF from the rumen should have priority in forage evaluation research.

Table 1. Kinetic characteristics of typical alfalfa and orchardgrass forages.

Kinetic Parameter	Alfalfa	Orchardgrass
NDF Content of the Forage	.50	.65
Fraction of NDF that is Potentially Digestible	.50	.70
Fraction of NDF ingested as large particles	.60	.70
Rate of NDF digestion	.085/h	.073/h
Rate of particle escape from the rumen	.04/h	.035/h
Rate of particle reduction and release	.07/h	.07/h
Observed dry matter intake	2.55%BW/d	2.28%BW/d
Observed dry matter digestibility	57.8%	61.6%

Table 2. Relationship of a 10% increase in kinetic characteristics on expected intake of ruminants.

Source of Characteristic	Kinetic Characteristic	% Change in Intake
Diet	NDF Content	−9.1
Diet	Potential Digestibility of NDF	+8.4
Animal	Rate of Ruminal Escape	+5.4
Diet/Animal	Size of Ingested NDF	−2.2
Diet	Rate of NDF Digestion	+2.2
Animal	Rate of Reduction/Release	+1.9

EFFECT OF SAMPLE SIZE AND RINSING PROCEDURE
ON NEUTRAL DETERGENT FIBER ANALYSIS

D.R. MERTENS

INTRODUCTION

Neutral detergent fiber has been used to develop a system for formulating dairy rations that maximizes dry matter intake and the proportion of the diet that is forage. The acceptance of NDF as a tool for formulating dairy ration by nutritional consultants and extension personnel in several states has stimulated the need for accurate and repeatable NDF analyses. Several modifications of the original method have been developed and many laboratories are making individual alterations in the procedure which may affect results. A series of experiments are planned to identify the critical steps in the NDF procedure and evaluate to the magnitude of errors created by modifications. The objective of this experiment was to evaluate the effect of rinsing method on NDF analyses. Two sample sizes were used to determine if there is an interaction between washing treatment and sample size.

MATERIALS AND METHODS

Forage samples were used to evaluate rinsing methods because they yield larger fiber residues than concentrates and have greater potential for retaining detergent and soluble matter with incomplete rinsing. Three legume samples with NDF concentrations of approximately 45, 50 and 55% and three grass samples with approximately 55, 70 and 80% NDF were selected to provide a range in NDF concentrations. Sample sizes of approximately .5 and 1.0 g dry matter were analyzed using a modification of the NDF procedure that uses amylase to remove starch

and eliminates the use of sodium sulfite and decahydronaphthalene. Refluxing time and method was identical for all samples. All residues were weighed hot immediately after removal from a 105°C oven to prevent moisture adsorption. Residue washing treatments included: zero rinse in which the sample was transferred into fritted-disk Gooch crucibles using 40 mls of boiling water with no rinses, two cycle rinse in which the residue was rinsed by adding 40 mls of boiling water and removing it by vacuum as rapidly as possible 2 times; four cycle rinse in which 4 rapid 40 ml rinses were used; two cycle soak in which 40 mls of boiling water was allowed to equilibrate with the residue for 5 minutes before vacuum was applied (repeated twice), three-cycle soak similar to 2S with three repetitions and four-cycle soak similar to 2S with four repetitions. Regular NDF, ash-free NDF and NDF ash were measured.

RESULTS AND DISCUSSION

Soaking the residue in boiling water resulted in lower NDF values than rapid rinsing. Rapid rinsing yielded lower values than zero washing (Table 1). It appears that rapid rinsing can remove 1.2% units of detergent and soluble matter that contaminates NDF residues, but soaking the residue in boiling water is needed to remove an additional .4% units of contamination within fiber spaces. Some of the contamination is soluble ash, which can be substracted if NDF is determined as ash-free NDF. Four

rapid rinses of the residue yielded ash-free NDF values that were not different from the values obtained by soaking the residue. Results indicate that soaking is needed to remove contamination from NDF residues and the maximum error attributed to detergent contamination of NDF residues is 1.6% units. The three grass samples averaged 2.1% units difference between zero rinse and soaking, while the three legumes averaged 1.2% units; however, the effect was not proportional to the NDF content of the forages. This suggests that thorough washing of residues is more critical in grass forage samples.

Reducing samples size from 1.0 to .5 g resulted in smaller NDF (61.7 vs. 61.0), smaller NDF ash-free (60.2 vs. 59.3) and larger NDF ash (1.5 vs. 1.7) concentrations. There was no sample size by treatment interaction indicating that the effect of sample size was constant across all washing treatments. There was a significant forage by sample size interaction. The difference between .5 and 1.0 g samples was 1.1, 1.2, 0.2, 0.4, 0.9 and 0.7 for mixed legume hay (46.5% NDF), alfalfa haylage (51.9% NDF), alfalfa hay (57.4% NDF), immature fescue (56.0% NDF), bromegrass hay (71.7% NDF) and wheat straw (81.9% NDF), respectively.

CONCLUSION

It is recommended that residues be soaked twice for approximately five minutes in order to remove detergent and soluble matter contamination from NDF residues and that further research be conducted to determine the effect of sample size on NDF analyses.

Table 1. Differences in neutral detergent fiber (NDF), ash-free NDF and NDF ash concentrations (% dry matter) related to residue washing treatment during fiber analysis.[1]

Residue washing treatment	NDF	Ash-free NDF	NDF ash
0 rinse	62.6[a]	60.6[a]	1.9[a]
2 rinse	61.6[b]	59.8[b]	1.8[ab]
4 rinse	61.3[b]	59.7[bc]	1.7[bc]
2 soak	60.9[c]	59.4[c]	1.5[cd]
3 soak	60.9[c]	59.4[c]	1.6[bcd]
4 soak	60.9[c]	59.5[bc]	1.4[d]

[1] Treatments with different superscripts differ (P<.05)

1986 U.S. DAIRY FORAGE RESEARCH CENTER FIELD FACILITY

AGRONOMIC REPORT

BRAD VENUTO

I. CROPS & ACREAGE

Crop	1986 Acres	1987 Acres
Alfalfa		
Established	319	330
New Seeding	89	153
Alfalfa/Ryegrass	30	0
Red Clover	5	5
Trefoil	5	5
Corn	464	298
Winter Wheat	35	55
Pasture		
Improved	5	5
Unimproved	275	275
Research	90	100
TOTAL	1317	1226*

*Approximately 90 acres of leased land was not renewed.

II. YIELDS

In 1986 we hauled 366 loads of haylage, 230 loads of hay, 87 loads of corn silage, and 109 loads of H.M. ear corn.

During the main harvest period comprising 120 days (May 20-September 1), we had 17 days of rain. We mowed forage on 44 days and harvested on 50 days.

D.M. yields per acre ranged from 3-6 tons per acre for alfalfa, 3.5-5.25 tons per acre for corn silage and 2.4-4.3 tons per acre for H.M. ear corn.

Crop	Tons D.M. harvested
Alfalfa haylage	858
Alfalfa hay	416
Corn silage	245
H.M. ear corn	610
Corn stover	38 (wet)

III. MANURE

Over 1900 loads of manure were hauled and applied in 1986. This consisted of approximately 4,000,000 gallons of liquid and 400 tons of solid. Total fertilizer value (N,P&K) approached $20,000.

IV. LIME AND FERTILIZER

760 tons of lime, 130 tons of alfala top-dress and starter, 36 tons of corn starter and 40 tons of anhydrous ammonia were applied in 1986.

1986 U.S. DAIRY FORAGE RESEARCH CENTER FIELD FACILITY
ANIMAL OPERATIONS REPORT
LEN L. STROZINSKI, DAIRY HERD MANAGER

Total livestock numbers now stand at 490 cattle and 52 sheep. Currently there are 215 cows milking, 35 cows dry and 240 herd replacements. We ship approximately 24,000 pounds of milk every other day. Our D.H.I.A. rolling herd average continues to rise slowly despite minimum culling pressure and now stands at 17,014 pounds of milk, 629 pounds of fat with a 3.7% test. Seventy percent of the milking herd are currently being used on research trials.

Our labor force now totals 18 full time employees: 13 experimental farm laborers, 1 maintenance mechanic, 1 secretary, 2 herd assistants and 1 herd supervisor. We operate with 2 working shifts (4:30 a.m.-1:00 p.m. and 1:30 p.m.-10:00 p.m.). One of the herd assistants has recently moved into the second residence at the farm.

This winter we were forced to abandon the facility's manure recycling system due to increased mechanical problems with the existing equipment and inability to handle the increased manure load. The equipment we had been using is no longer manufactured. Replacement parts are difficult to come by and are expensive. We have evaluated other manure separator systems but have not found a unit which we feel will meet our needs. Therefore, Dr. Koegel and his staff are redesigning our old system. In the mean time, we are bedding with straw and sawdust and the manure is being handled as slurry manure.

I have just completed transfer of all our dairy cattle records to a new computer system. The new system, Dairy Comp 305 by Valley Agricultural Software, runs on IBM compatible equipment. It is much faster than the old system and provides the ability to easily transfer data directly to the researcher's computer.

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